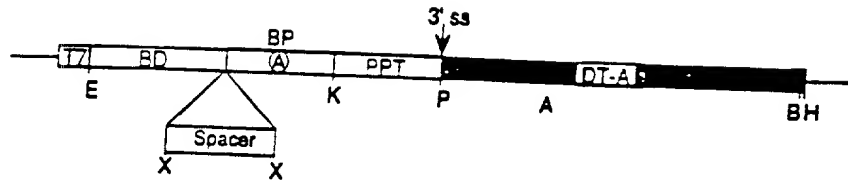


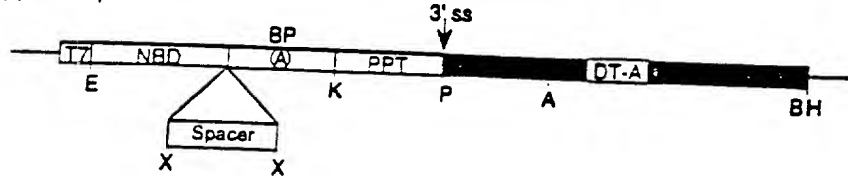
FIGURE 1A



(B)



(2) pPTM-Sp



(C)

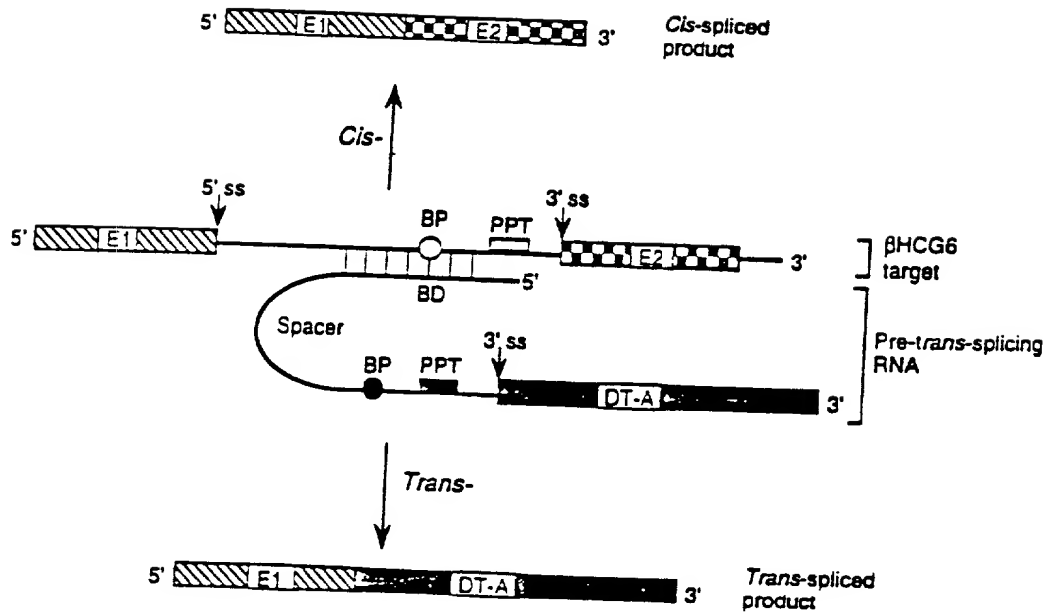
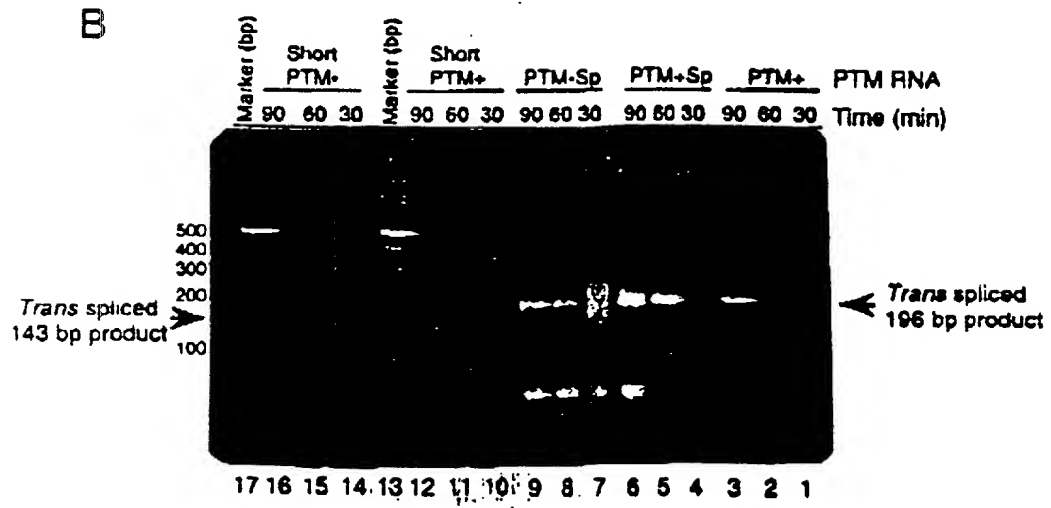
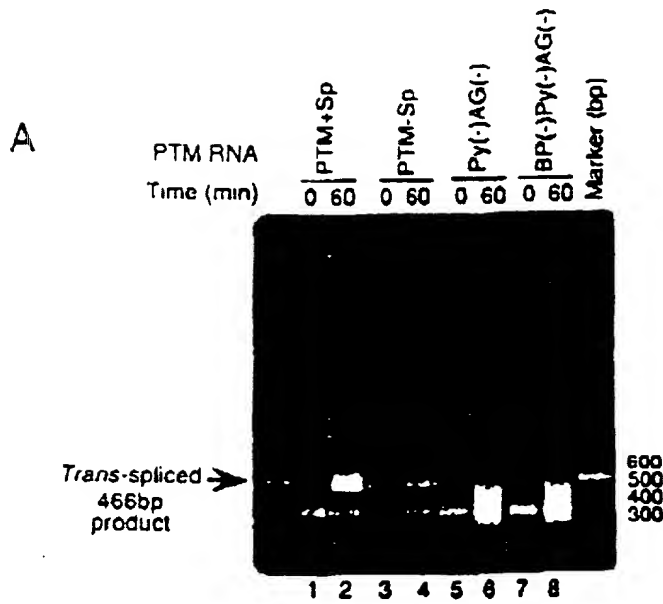
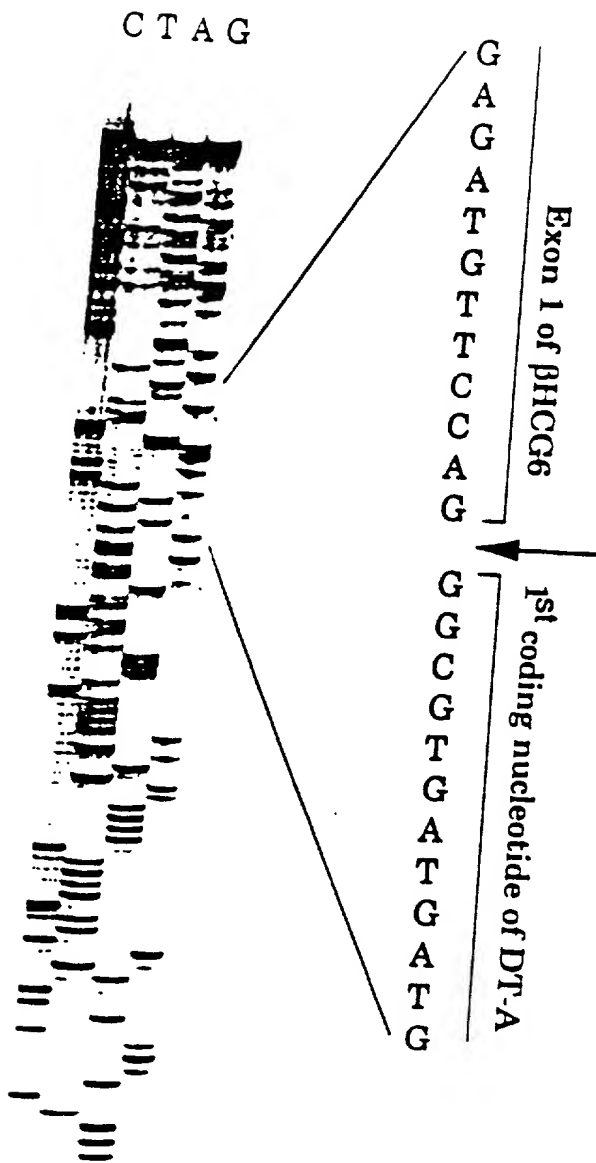


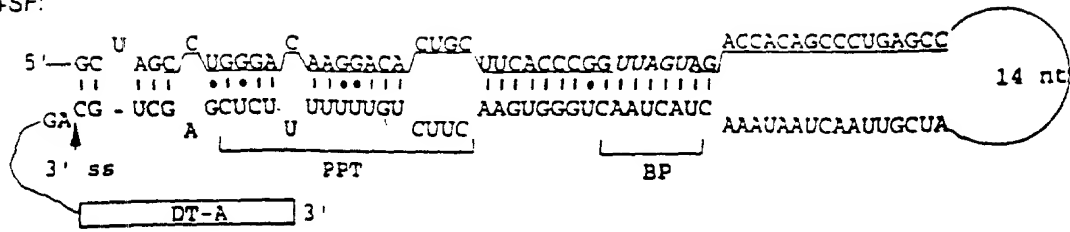
Figure 1 B-C



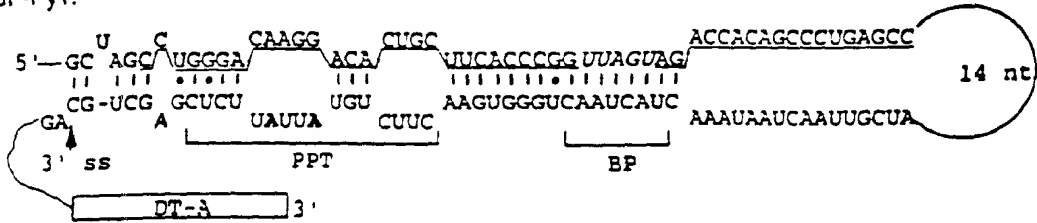


(A)

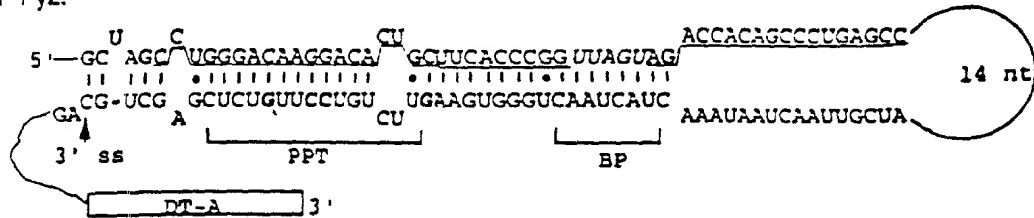
1. PTM+SF:



2. PTM+SF-Py1:



3. PTM+SF-Py2:



(B)

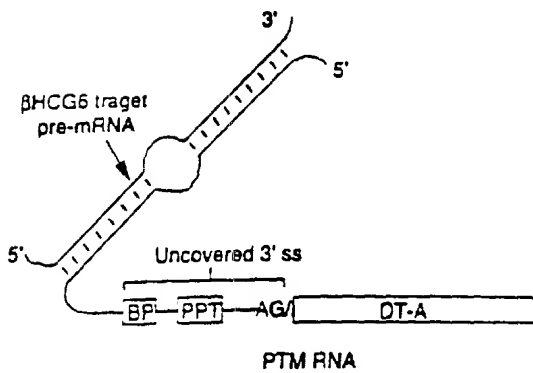


Figure 4A-B

(C)

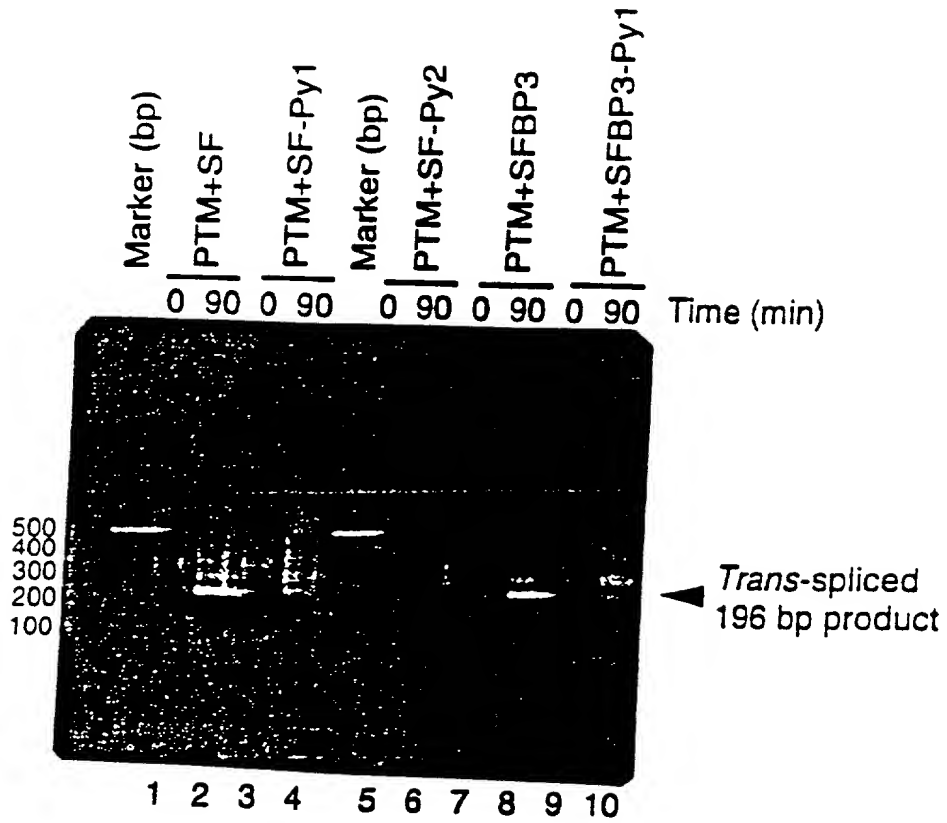


Figure 4c

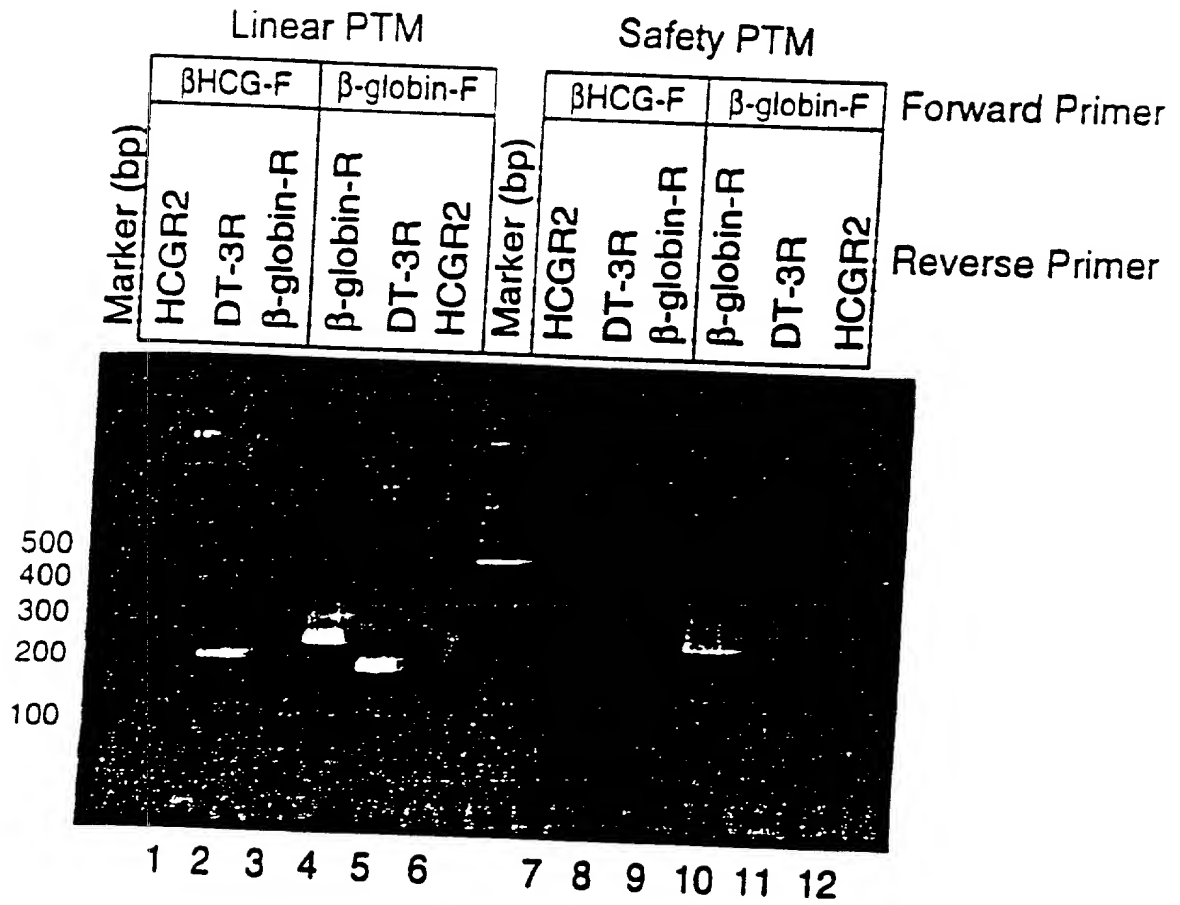


Figure 5

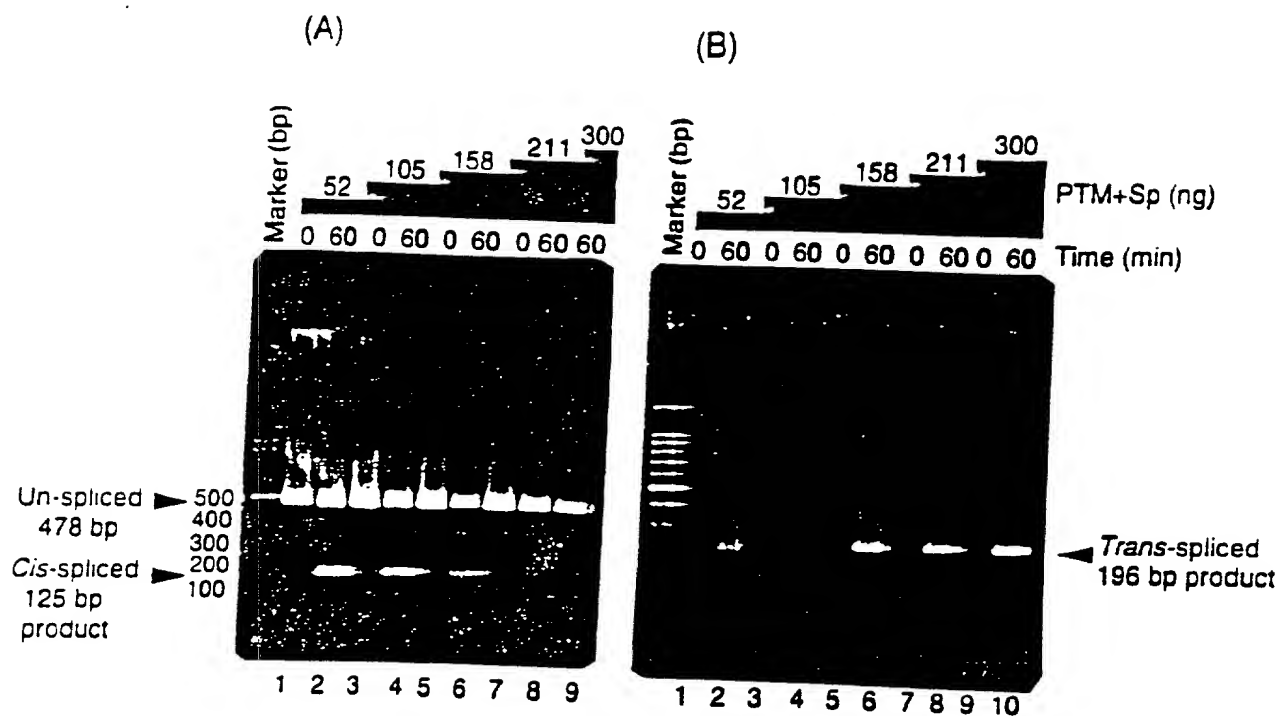
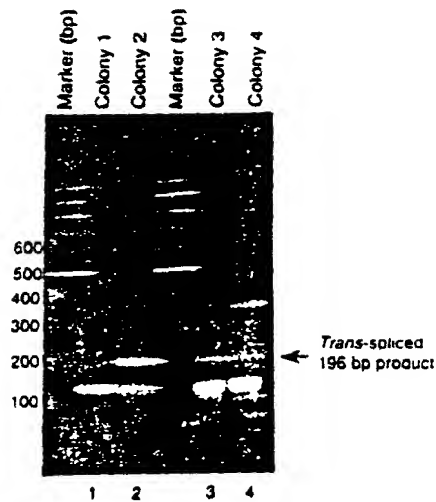


Figure 6

Figure 7

(A)



(B)

Exon 1 of β HCG6 ↓
 5'-CAGGGGACGCACCAAGGATGGAGATGTTCCAG-GGCGCTGATGATGTTGTT
 ↑ 1st coding nucleotide of DT-A
 GATTCTTCTTAAATCTTTTGTGATGGAAAACCTTTTCTTCGTACCACGGGACTA
 AACCTGGTTATGTAGATTCCATTCAAAA-3'

Double Splicing Pre-therapeutic RNA

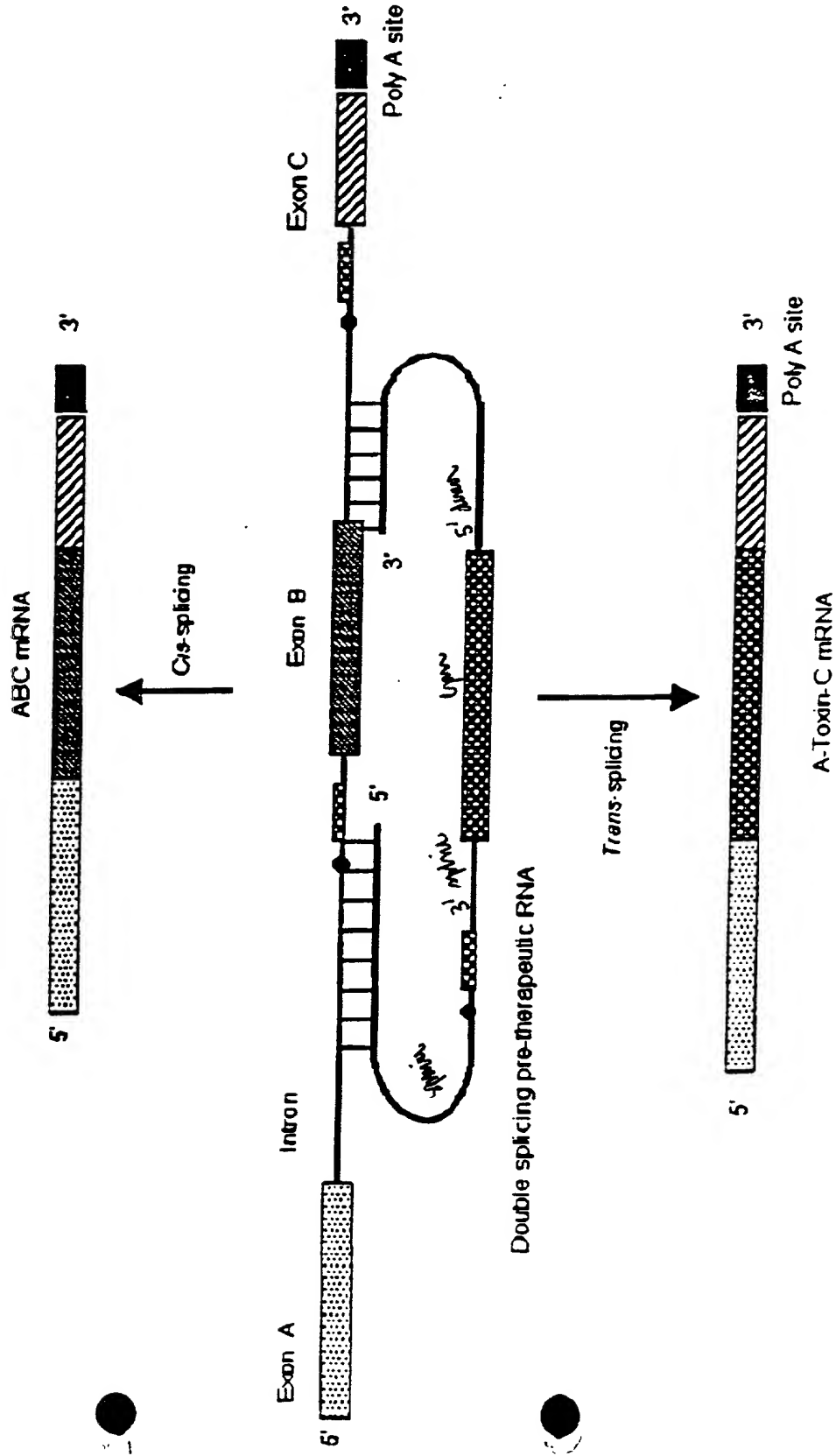


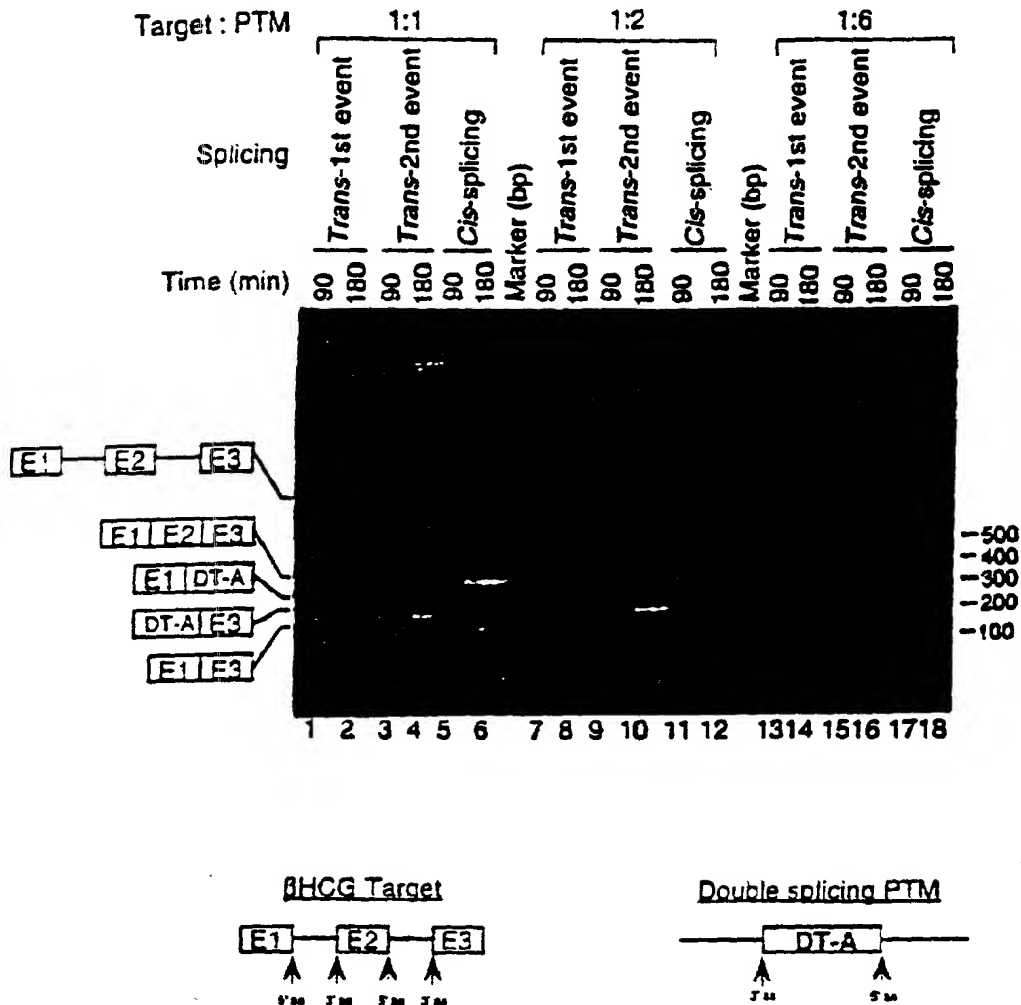
Figure 8 A

31304B-A

(Sheet 12 of 58)

Selective Trans-splicing of a Double Splicing PTM

(3' ss of PTM to 5' ss target and, 5' ss of PTM to 3' ss of target)



Cis-spliced products

E1 E2 E3 = Normal cis-splicing (277bp)

E1 E3 = Exon skipping (110bp)

Trans-spliced products

E1 DT-A = 1st event, 196bp. Trans-splicing between 5' ss of target & 3' ss of PTM.

DT-A E3 = 2nd event, 161bp. Trans-splicing between 3' ss of target & 5' ss of PTM.

Figure 8B

31304B -A

(Sheet || Of 58)

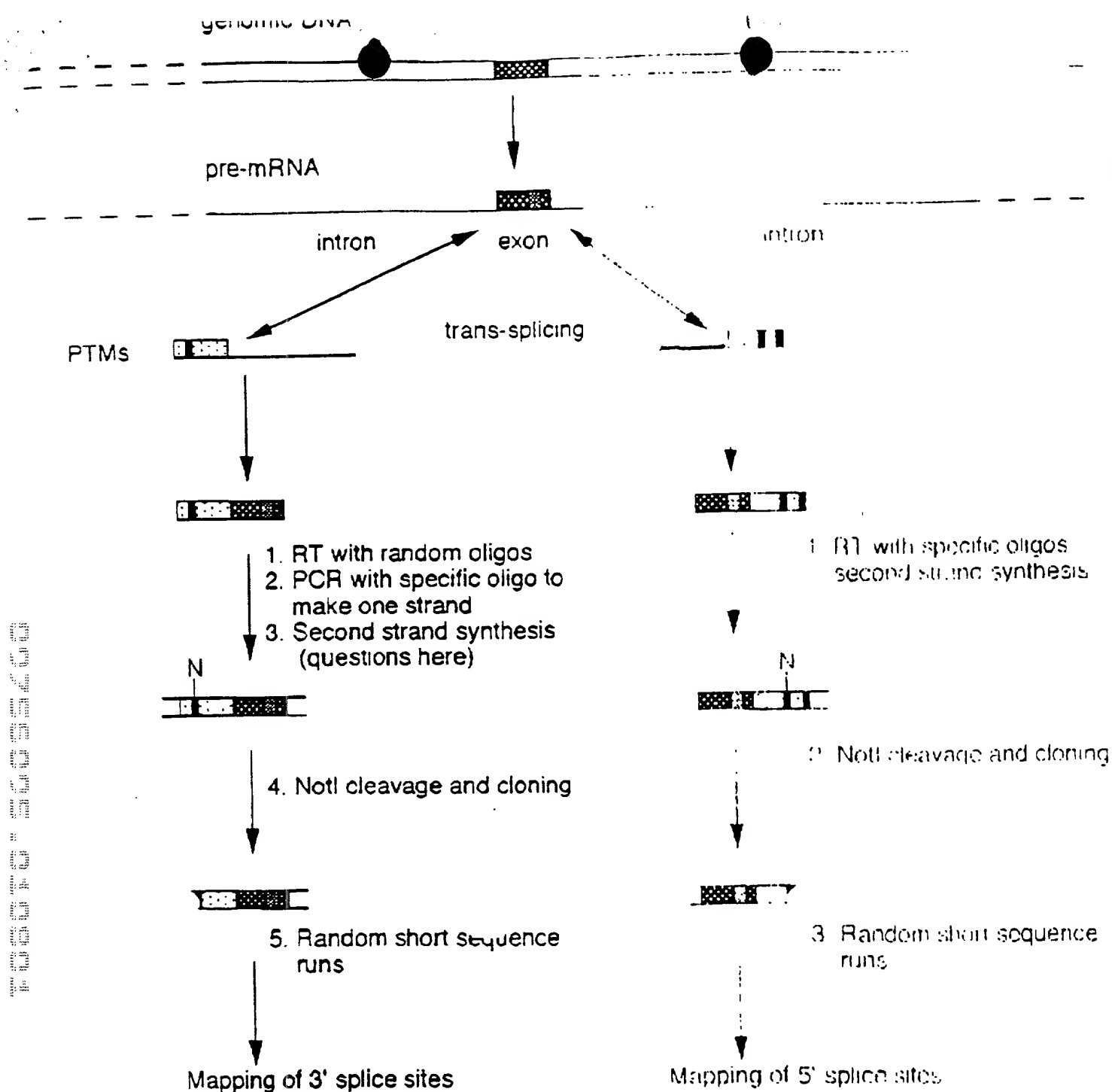


FIGURE 9

31304B-A
(Sheet 12 Of 58)

[illegible]

Knock Out

Target 1:

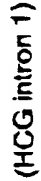


04 HC 5

2/

9

pc 8.1 PTM2 :-



Restoration of β -Gal activity by SMaRT

(Spliceosome Mediated RNA *Trans*-splicing)

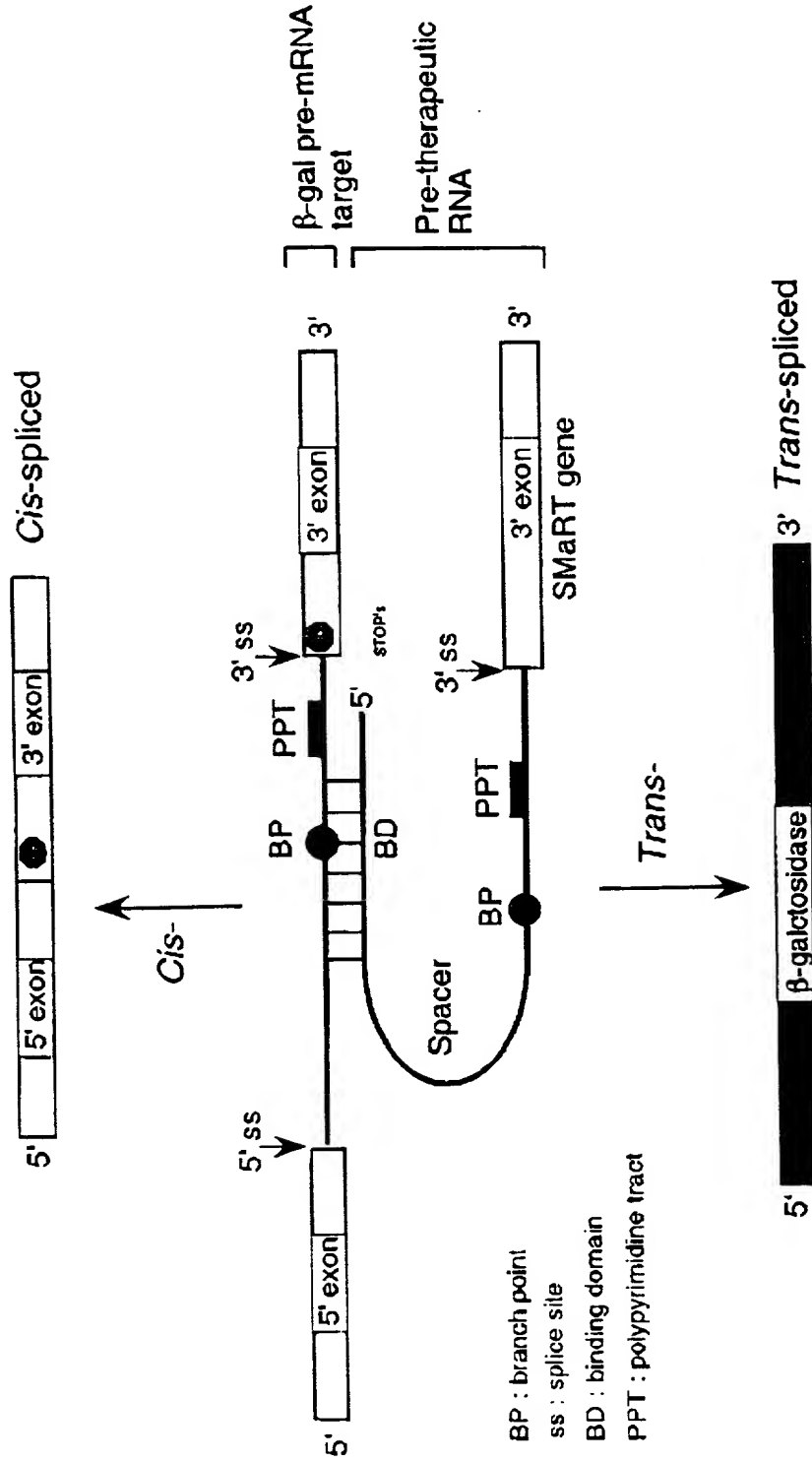


Figure 10B

31304 B-A

(Sheet 14 of 21)

31304 B-A
(Sheet 15 of 58)

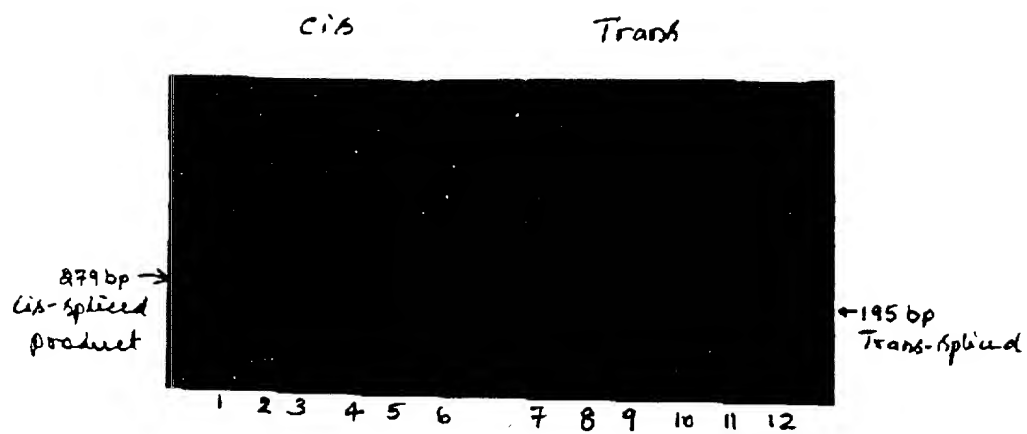


FIGURE 11A

31304 B-A
(Sheet 16 of 58)

Figure 11B

51507 15-11
(Sheet 17 of 58)

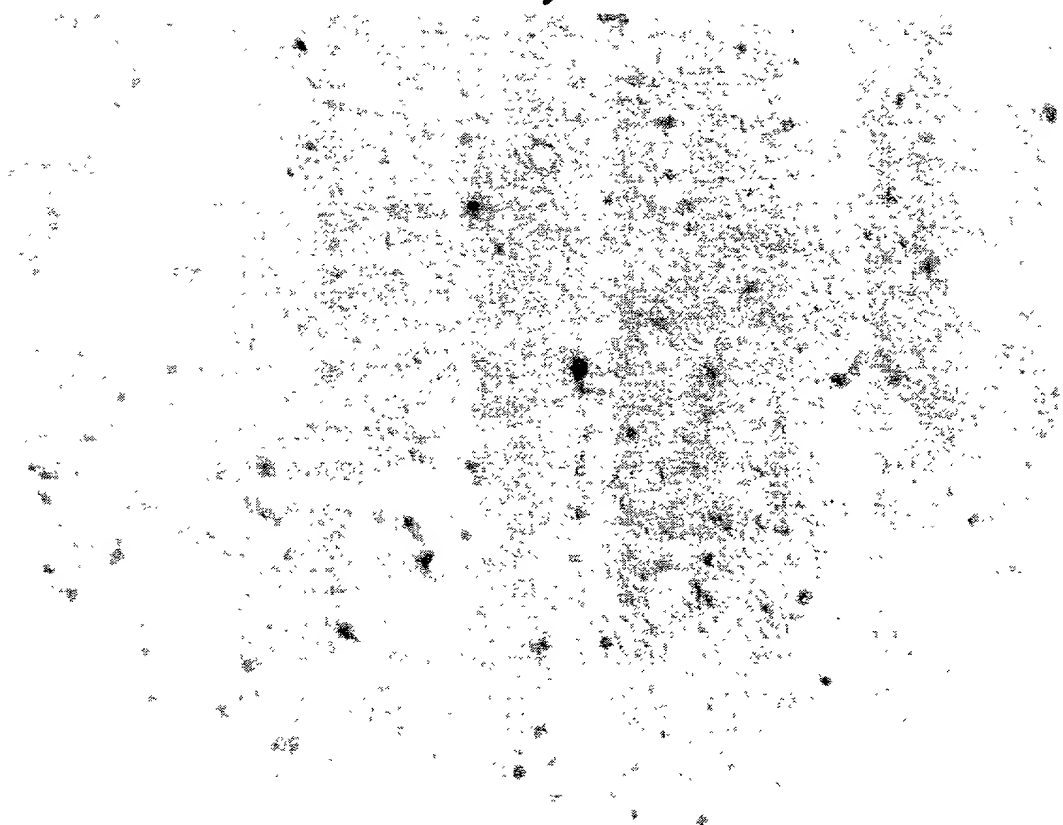


FIGURE 11C

Nucleotide Sequence Demonstrating that *Trans*-splicing is Accurate

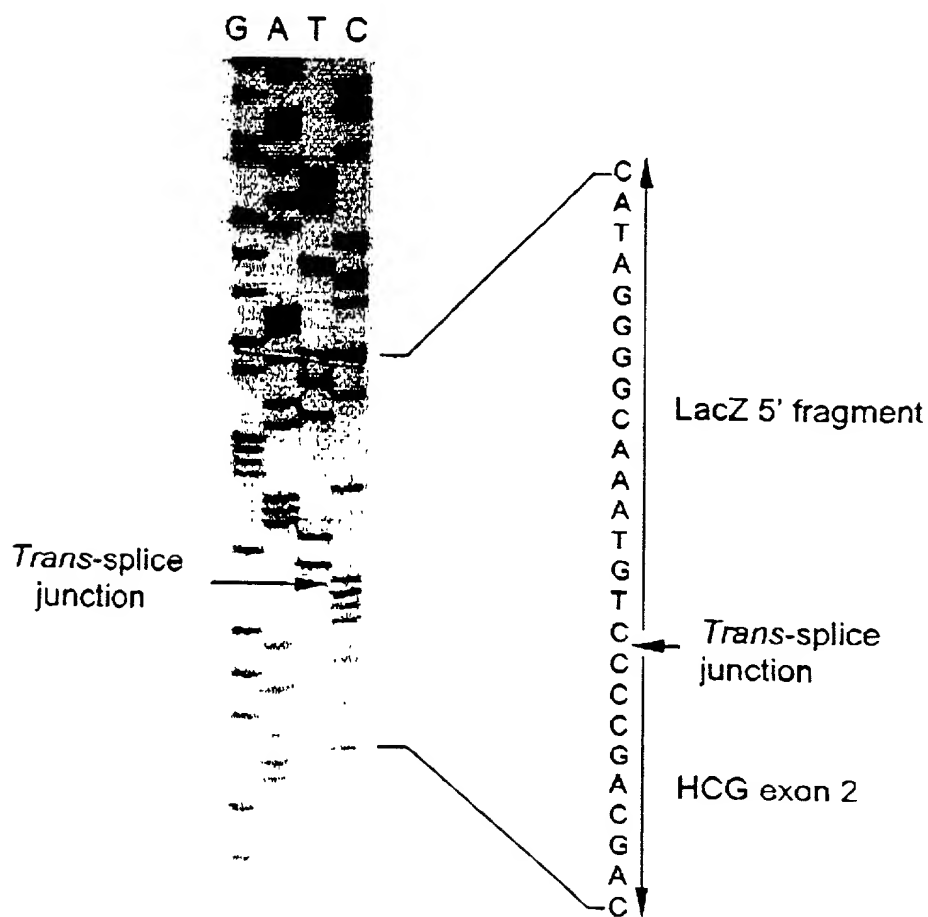


FIGURE 12 A

31304-B-A
(Sheet 18 of 58)

(1). Nucleotide sequences of the cis-spliced product (285 bp) :

BioLac-TR1

GGCTTTTCGCTACCTGGAGAGACGCGCCCGCTGATCCTTTGCGAATACGCCCACGCGATGGGTAAACAGTCTTG

Splice junction

CGGTTTCGCTAAATACTGGCAGGCGTTTCGTCAAGTATCCCCGTTTACAG/GGCGGCTTCGTCTAAATAATG

GGACTGGGTGGATCAGTCGCTGATTAAATATGATGAAAACGGCAACCCGTGGTGGCTTACGGCGGTGATT

Lac-TR2

TGGCGATACGCCGAACGATCGCCAGTTCTGTATGAACGGTCTGGTCTTTGGCGACCGCACGCCGCATCCAG

(2) Nucleotide sequences of the trans-spliced product (195 bp)

BioLac-TR1

GGCTTTTCGCTACCTGGAGAGACGCGCCCGCTGATCCTTTGCGAATACGCCCACGCGATGGGTAAACAGTCTTGG

Splice junction

CGGTTTCGCTAAATACTGGCAGGCGTTTCGTCAAGTATCCCCGTTTACAG/GGGCTGCTGCTGTTGCTGCTGCT

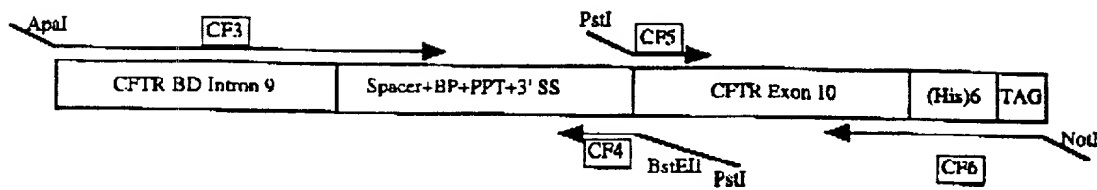
HCGR2

GAGCATGGGCGGGACATGGGCATCCAAGGAGCCACTTCGGCCACGGTGCCG

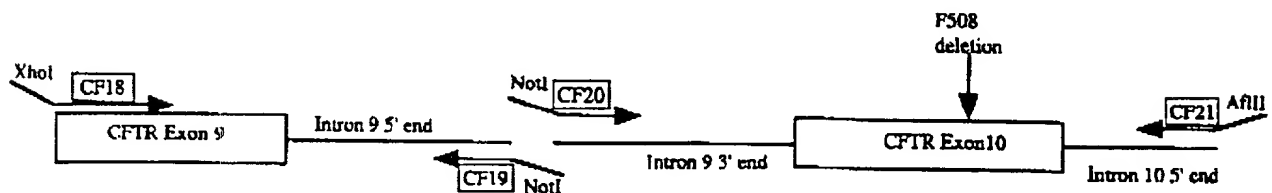
Figure 12 B

31304-B-A
(Shut 19 of 58)

CFTR Pre-therapeutic molecule (PTM or "bullet")



CFTR mini-gene target - Construction



TRANS-SPLICING Repair

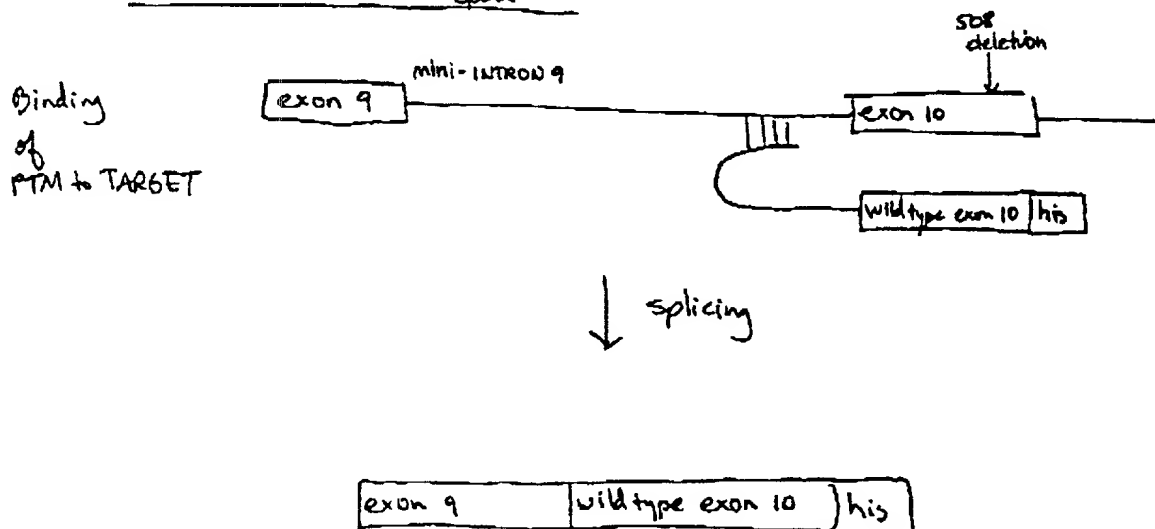
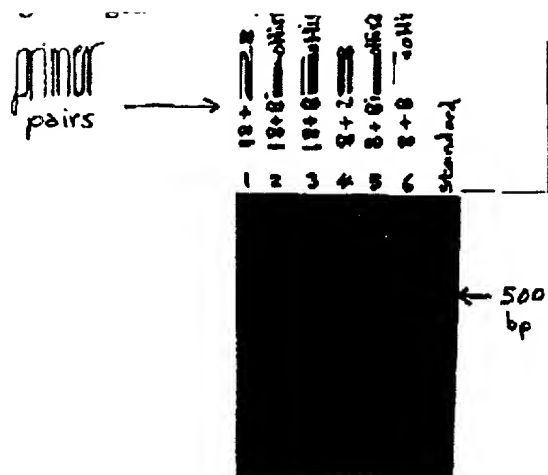


Figure 13

31304-B-A
(shut 2004.58)

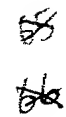
Figure 14



31304 B-A
(Sheet 21 of 58)

1

1



55
56

Acc I	-	EcoR I	1	Nde I	-	Sau96 I	2
Apa I	1	EcoR V	1	Nhe I	1	Sca I	1
ApaL I	-	Hae II	-	Not I	1	Sma I	-
Avr II	-	Hae III	2	PflM I	-	Sph I	1
BamH I	1	HinC II	-	Pst I	2	Spl I	-
Ban II	2	HinD III	1	Pvu I	-	Ssp I	-
Bbe I	-	Hinf I	-	Pvu II	-	Stu I	-

31304-A-B
(Ahtut 2201.58)

EXPERIMENT 2

Repair of an exogenously supplied CFTR target molecule carrying an F508 deletion in exon 10.

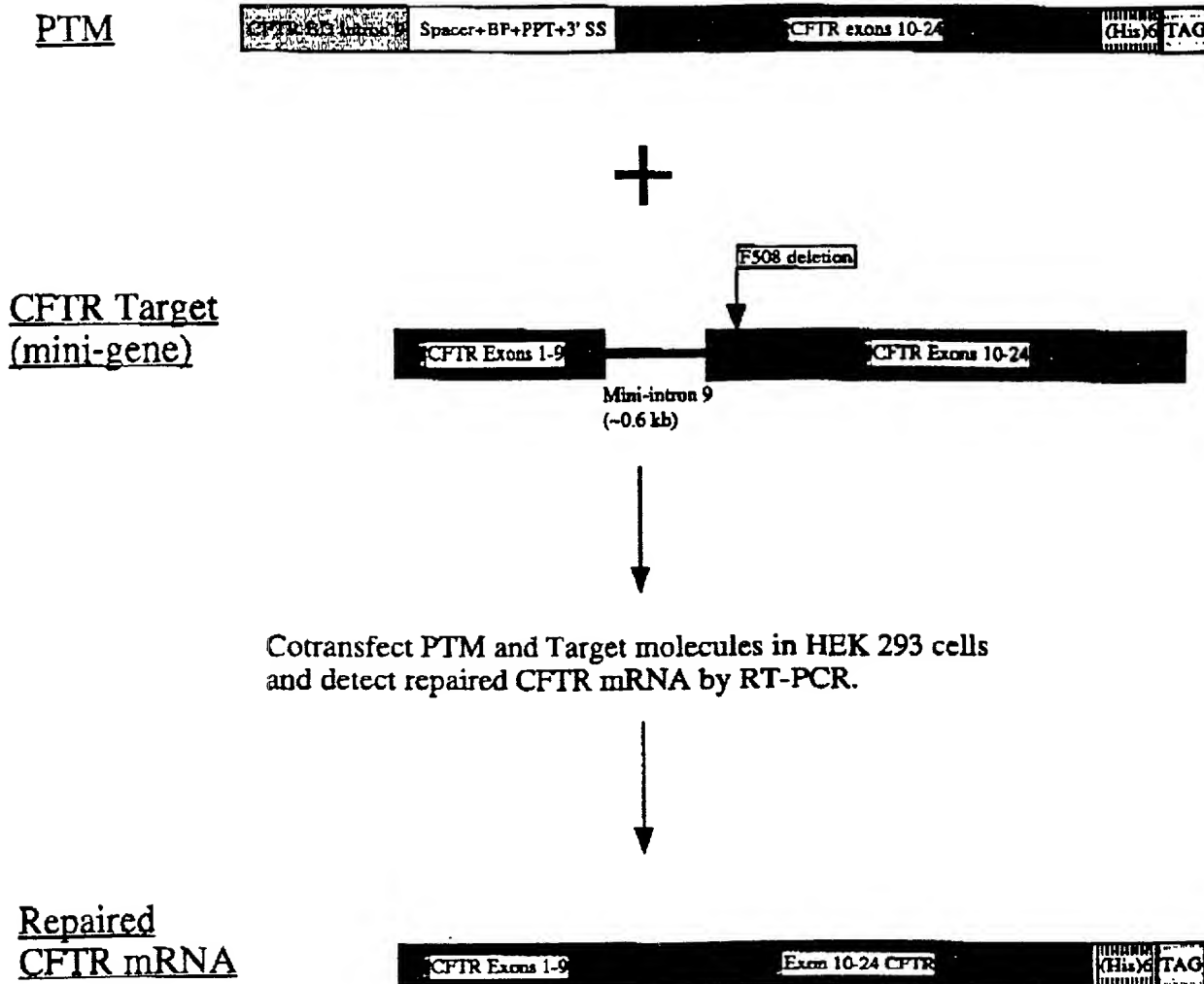


Figure 1b

31304-A-B

Sheet 23 of 50

EXPERIMENT 3

Repair of endogenous CFTR
transcripts by exon 10 invasion
using a double splicing PTM

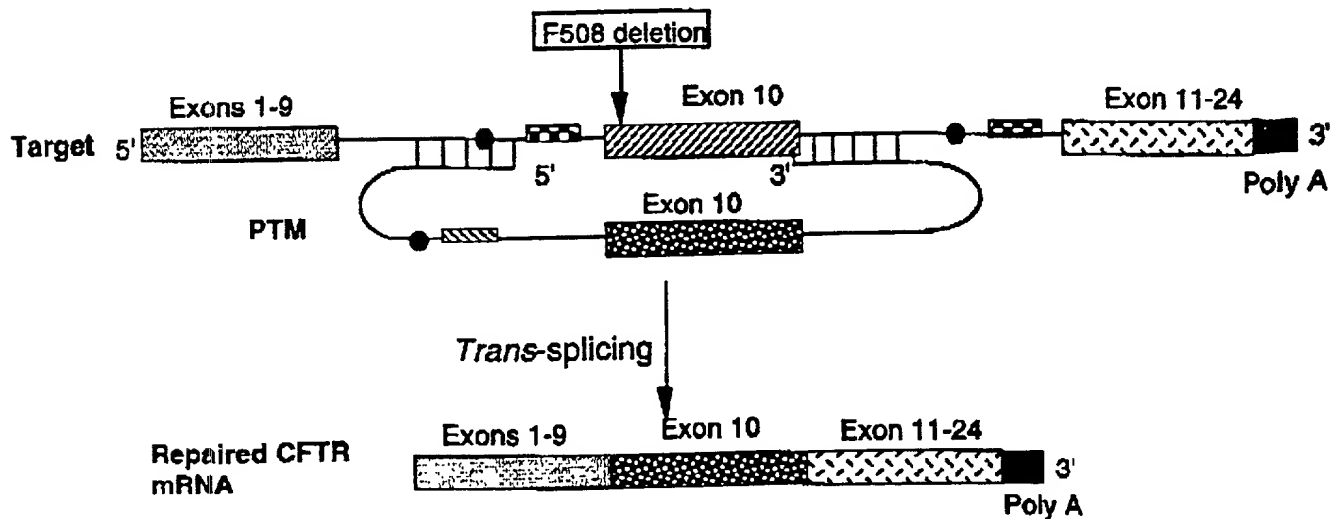
Double Splicing
PTM

Figure 17

31304 B-A

Sheet 24 of 58

Double Trans-splicing Specific Target

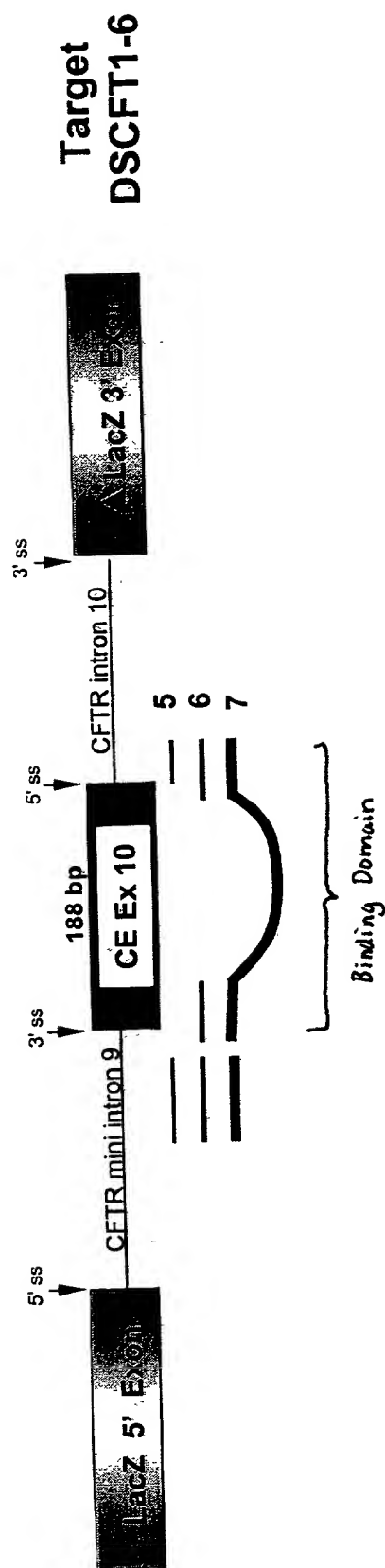
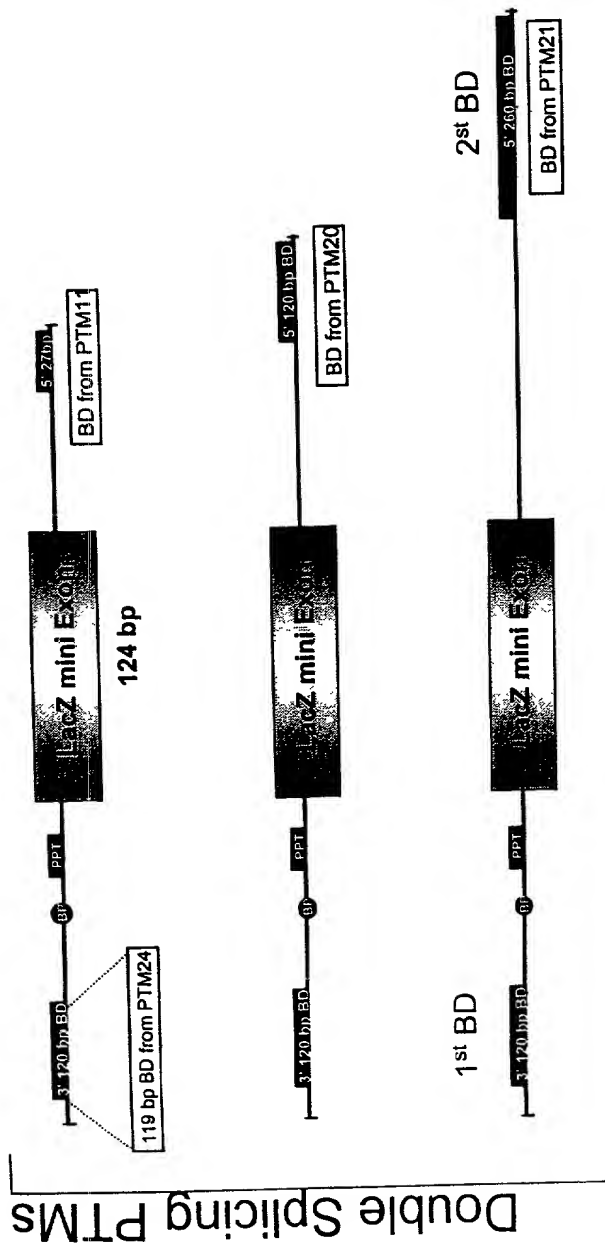


Figure 18

Double Trans-splicing PTMs



- DSPTM-5**
PTM with 27 bp BD & masks 5' single splice site
- DSPTM-6**
PTM with 120 bp BD & masks both 5' & 3' splice sites
- DSPTM-7**
PTM with 260 bp BD masking both the ss & the entire CFTR Ex10

Figure 19

Sheet 27 of 58

Double Trans-splicing β -Gal Model

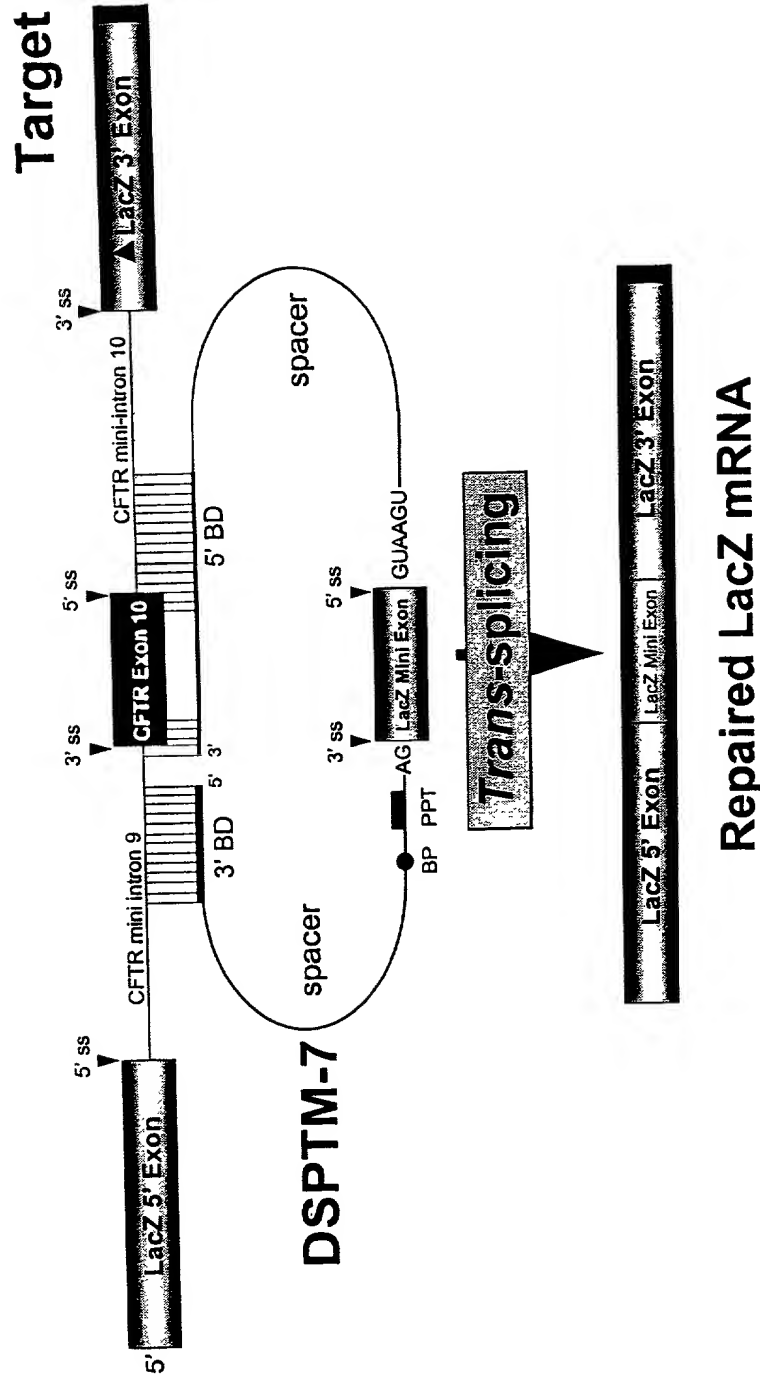
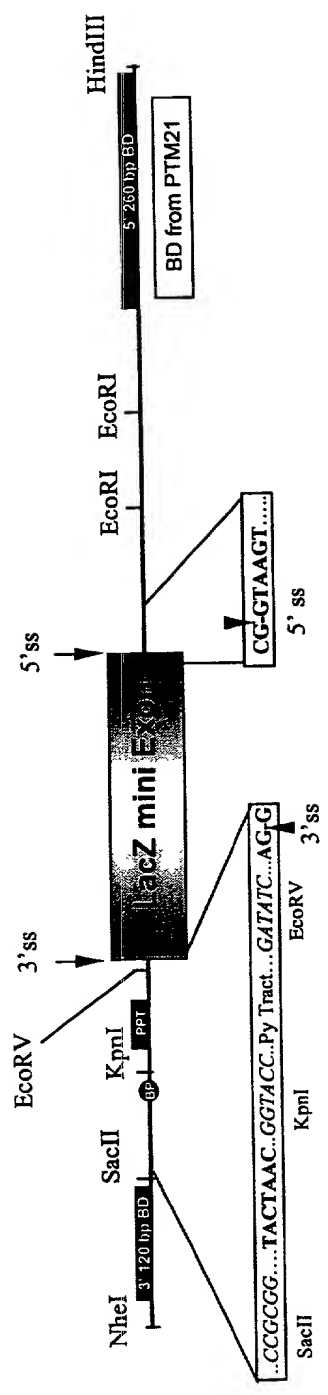


Figure 20

Important Structural Elements of DSPTM-7: (Double splicing PTM with all the necessary splice elements i.e. has both 3' and 5' functional splice sites and the binding domains)



(1) 3' BD (120 BP) : GATTCACCTTGCTCCAAATTATCATCCTAAGCAGAAGTGATATTTATTGTAAAGATTCTATTAACTCATTTGATTC
 AAAATATTTAAATACTTCTGTTTCATACCTGCTATGCAC

(2) Spacer sequences (24 bp): AACATTATTATAACGTTGCTCGAA

(3) Branch point, pyrimidine tract and acceptor splice site: TACTAAC T GGTAAC TCTTCTTTTTTTTTT GATATC CTGCAG **GAG GGC**
 3' ss Kpn I PPT EcoRV LacZ mini exon

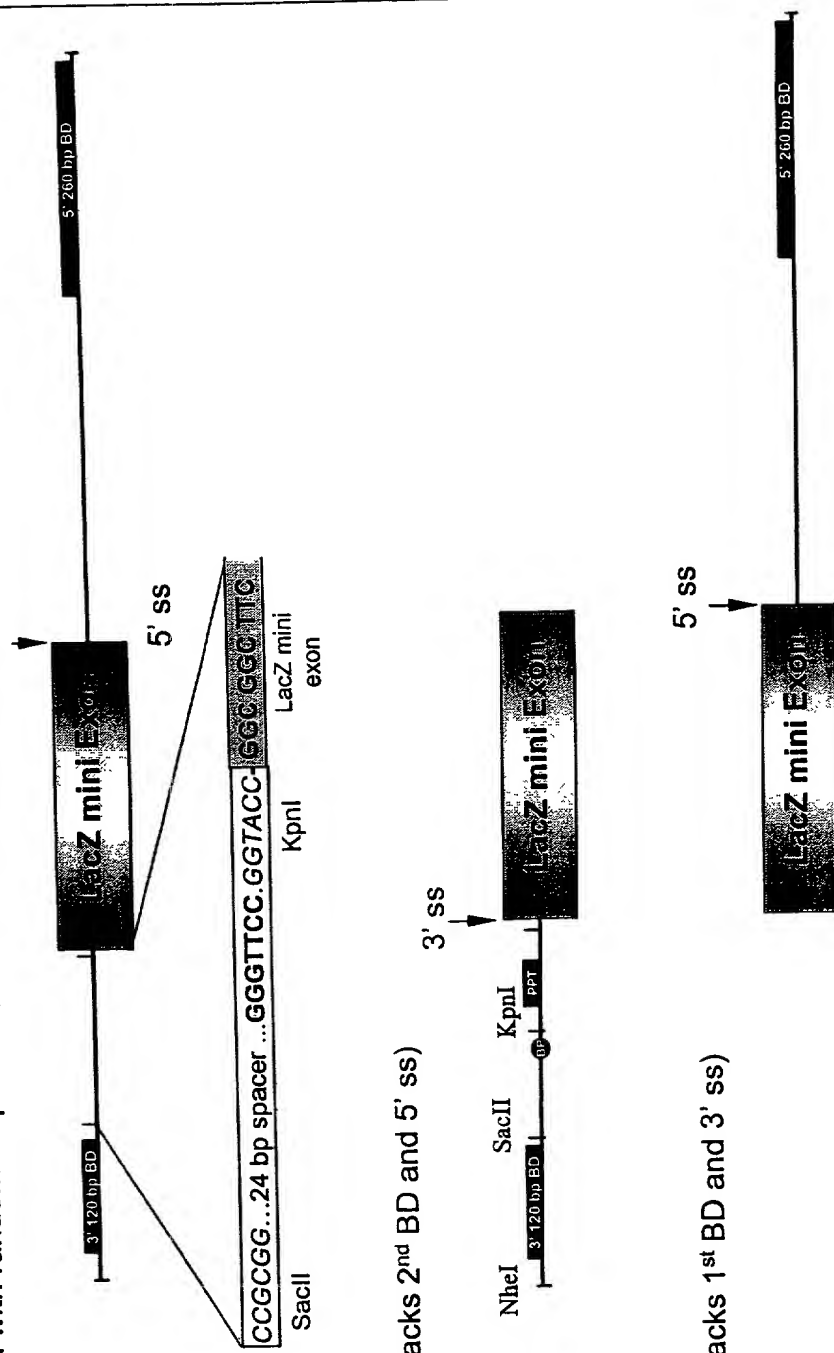
(4) 5' donor site and 2nd spacer sequence: **TGA ACG** GTAAGT GTTATCACCGGATAGTGTCTAACCTGATTCGGGCCCTTCGATACG
 5' ss LacZ mini exon

CTAAGATCCACCGG

(5) 5' BD (260 BP) : TCAAAAAGTTTTACATAATTTCTTACCTCTTCTTGAATTCATGCTTTGATGACGCTTCTGTATCTATATTCATCTGGAA
 ACACCAATGATTTTCTTTAATGGTGCCTGGCATAATCCTGGAAACTGATAACACAATGAAATTTCTCCACTGTGCTTAA
 AAAACCCCTCTGAAATTCCTCAATTTCTCCATAATCATCATTAACAACTGAACCTGTGGAATAAAACCCATCATTTATTAACTCA
 TTATCAAATCACGC

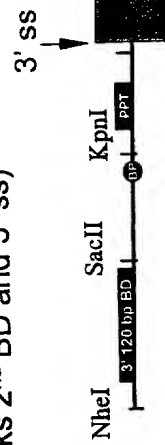
Figure 21

DSPTM8 : (▲ 3' ss: 3' splice elements i.e. BP, PPT & AG dinucleotide has been deleted and replaced with random sequences, but still has the functional 5' splice site)



Mutants

PTM29 (lacks 2nd BD and 5' ss)



PTM30 (lacks 1st BD and 3' ss)



Figure 22

Accuracy of Double Trans-splicing Reaction

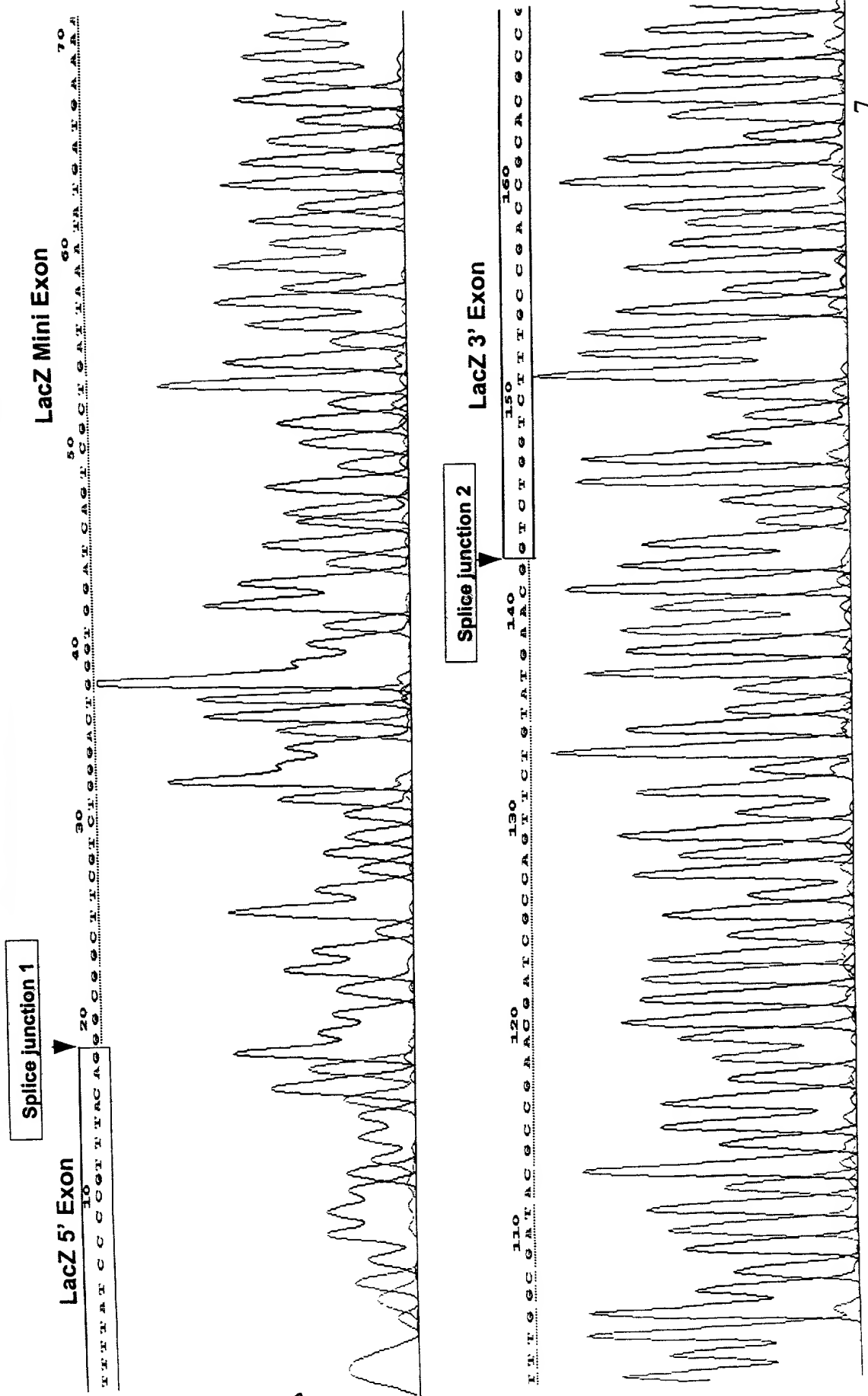


Figure 23

Sheet 31 of 58

Double Trans-splicing Produces Full-length Protein



1 2 3 4 5 6 7

Lane 1: DSCFT1.6 Target alone 25 µg
Lane 2: DSPTM7 25 µg
Lane 3 Target + PTM #6 25 µg
Lane 4: Target + PTM #9 25 µg
Lane 5: Delta 3' splice mutant alone 25 µg
Lane 6: Target + Delta 3' ss 25 µg
Lane 7: Target+PTM29+30 (mutants) 25 µg

Figure 24

Sheet 32 of 58

Restoration of β -Gal Function by Double Trans-splicing

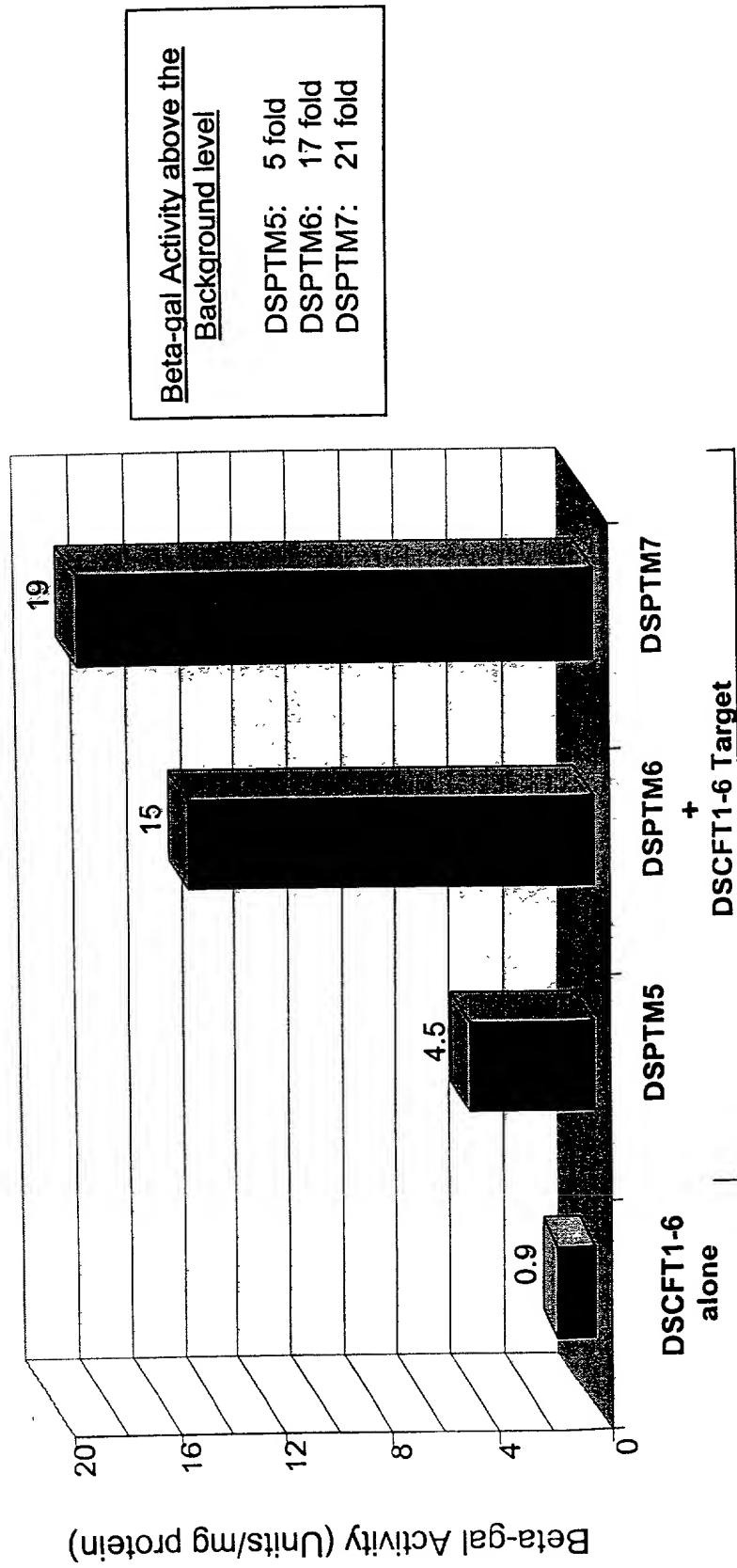


Figure 25

Restoration of β -gal activity is due to double RNA trans-splicing events

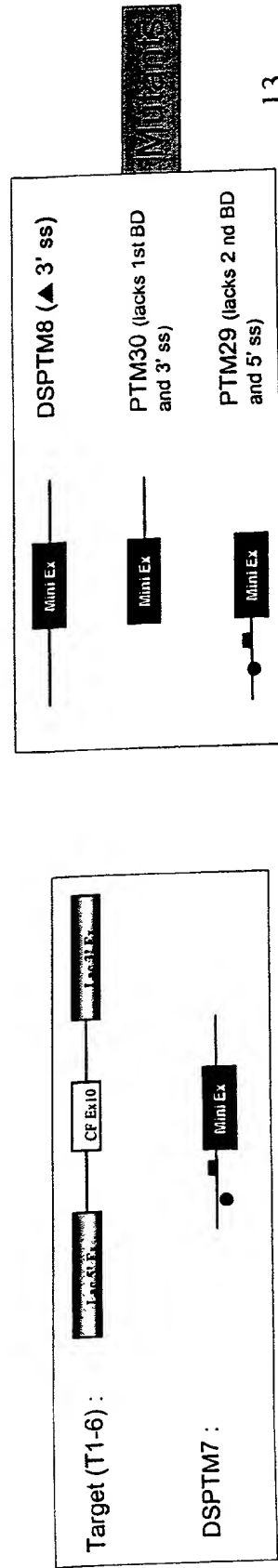
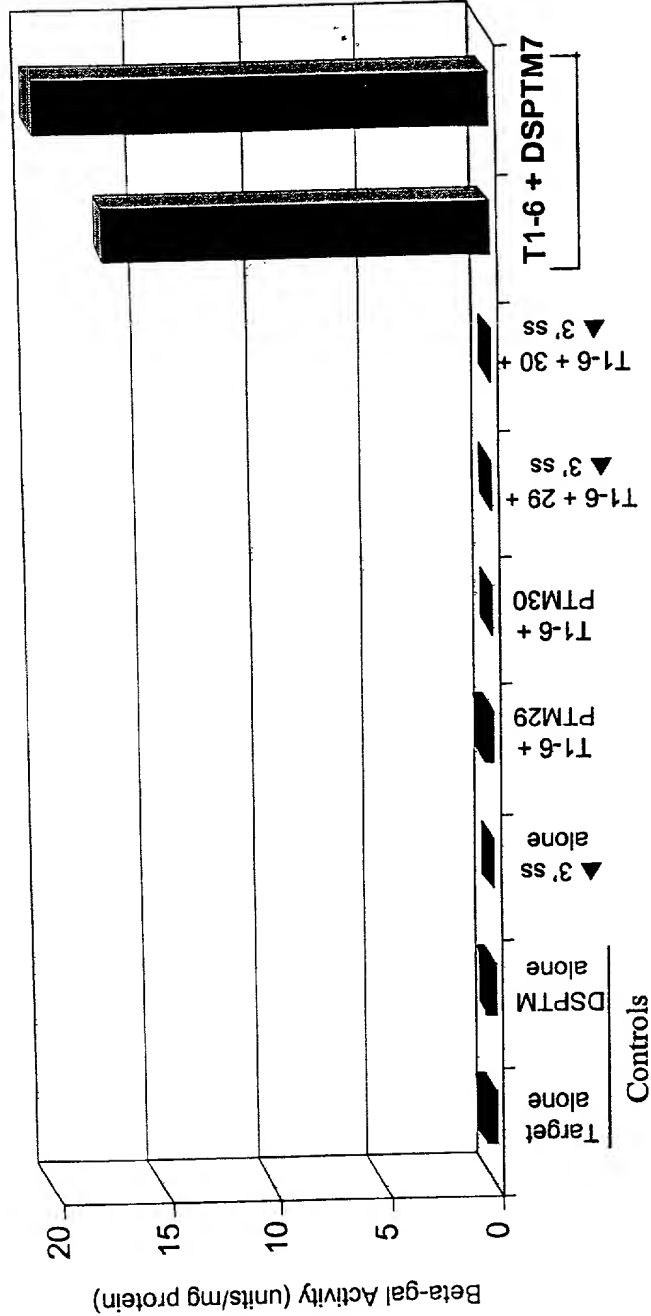
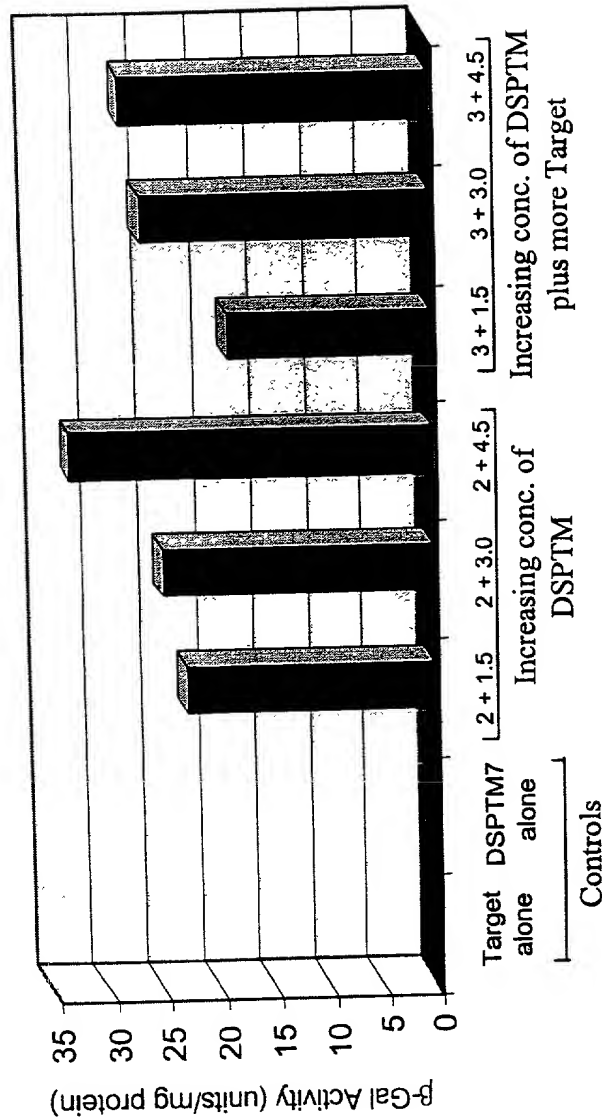


Figure 26

Sheet 34 of 58

Double Trans-splicing: Titration of Target & PTM

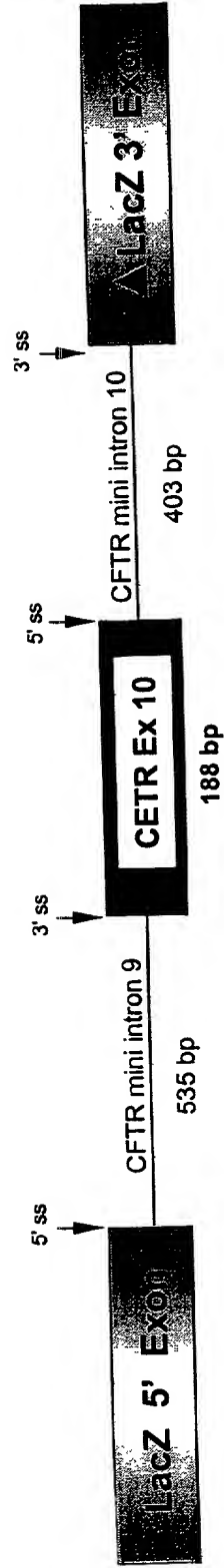


The current level of beta-gal activity due to double trans-splicing is ~ 1-1.5% of the best single splice model (3' exon replacement)

Figure 27

Sheet 35 of 58

DSCFT1-6 (Specific Target):



DSHCGT1 (Non-specific Target):

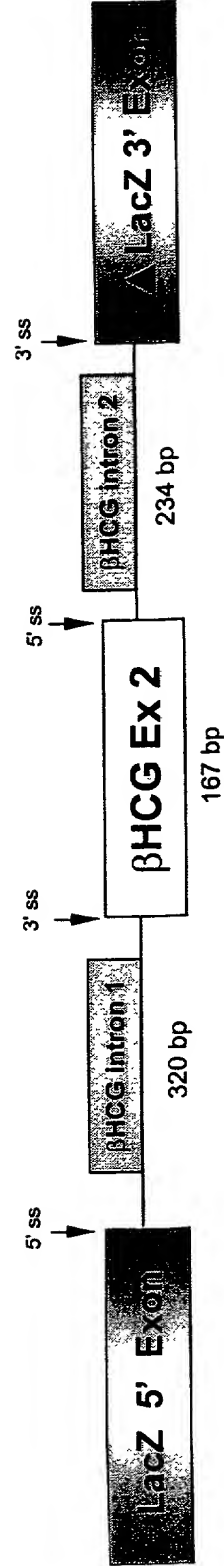


Figure 28

Specificity of double *trans*-splicing Reaction

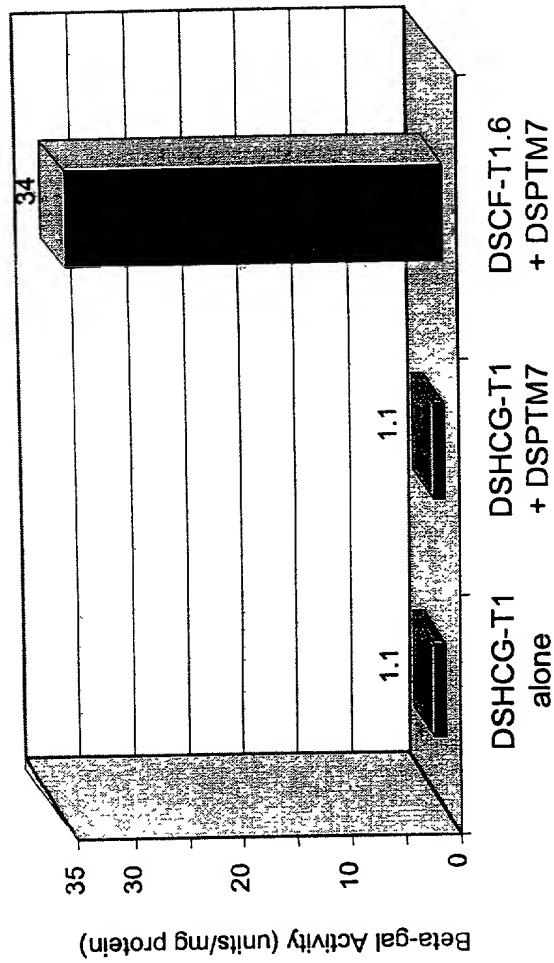
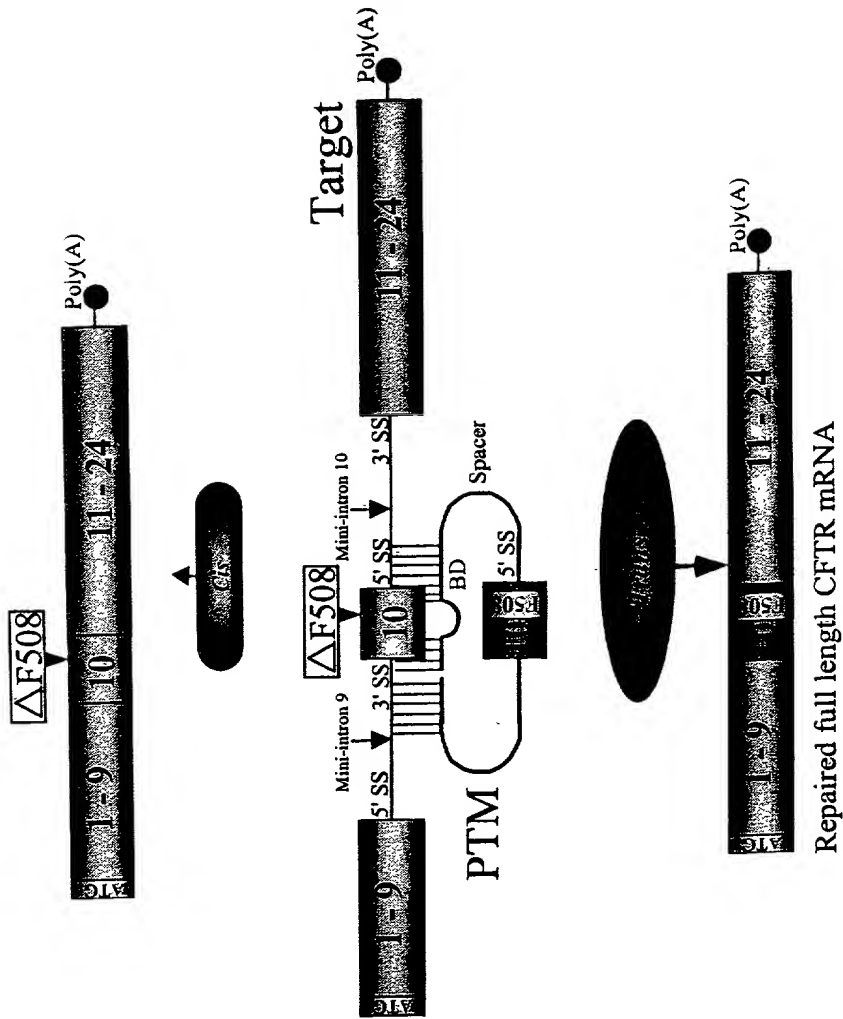


Figure 29

Sheet 37 of 58

Replacement of a Single Internal Exon Schematic diagram of a PTM binding to a CFTR ΔF508 target



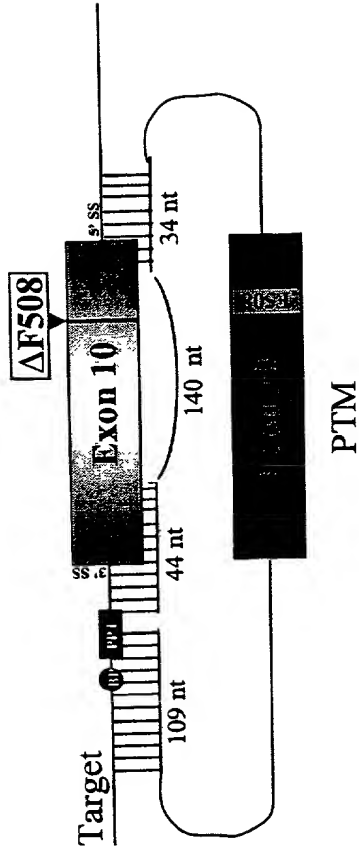
INTRONIN

Figure 30

about 38 of 58

the first 38 of 58

PTM with a long binding domain masking two splice sites and part of exon 10 in a mini-gene target.



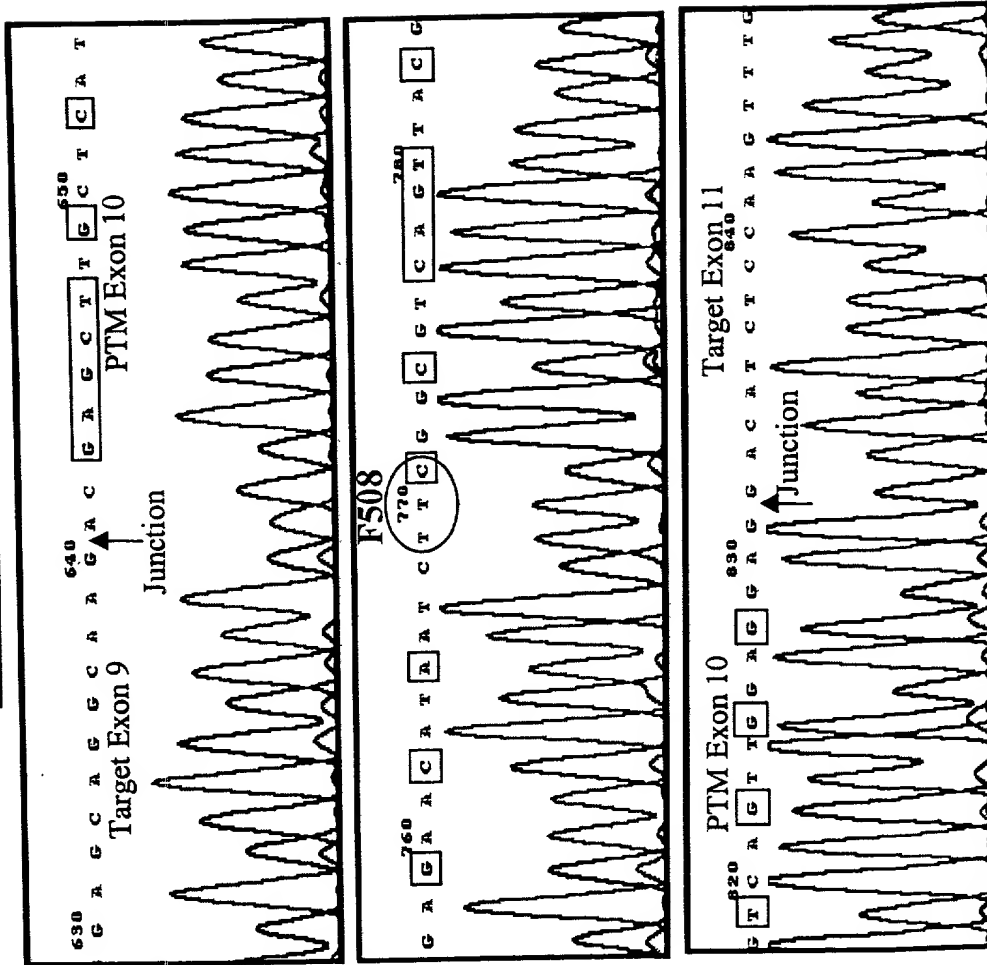
ACGAGCCTTGCTCATGATGATCATGGCGAGTTAGAACCAAGTGAAGGCAAGATCAAACATTCCCG
 GCCGCATCAGCTTTTGCAGCCAAATTCAGTTGGATCATGCCCGGTACCATCAAGGAGAAACATAAT
 CTTCCGCCGTCAGTTACGACGAGTACCGGTATCCGCTCGGTGATTAAGGCCCTGTCAGTTGGAGGAG

MCU in exon 10 of PTM
 88 of 192 (46%) bases in PTM exon 10 are not complementary to
 its binding domain (bold and underlined).

Figure 31

INTRON

Sequence of a double
trans-spliced product



□ = MCU in
PTM exon 10

Figure 32

about 40 of 58

CFTR Repair: 5' Exon Replacement

Schematic diagram of a PTM binding to the splice site of intron 10 of a mini-gene target

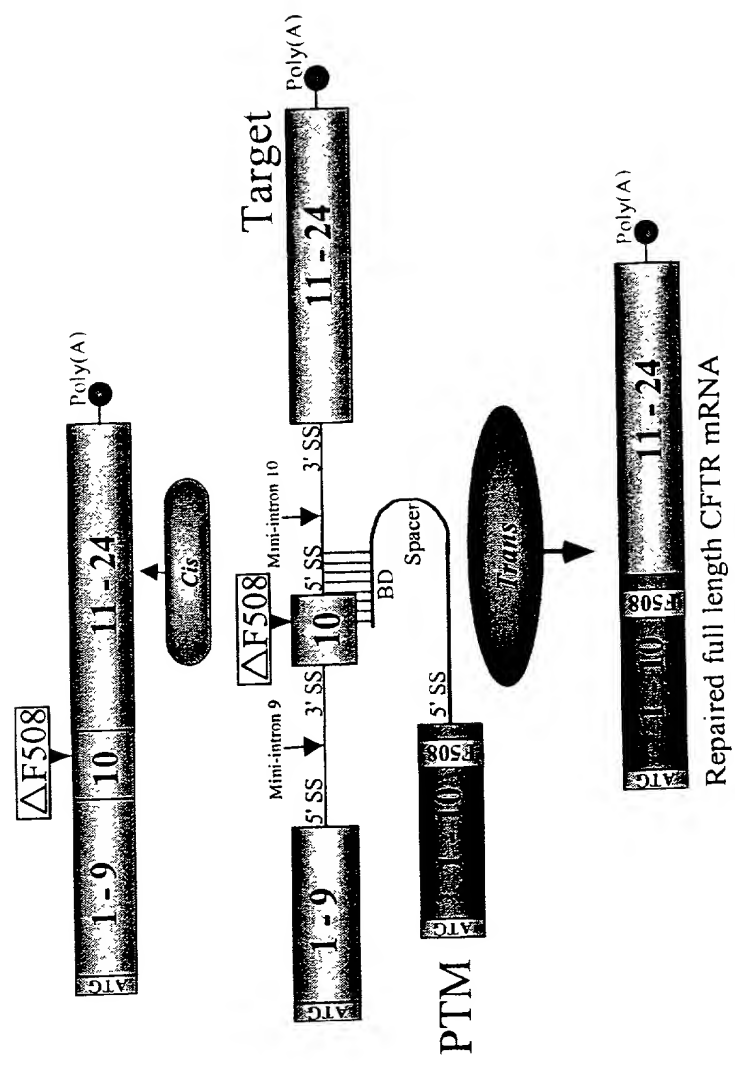
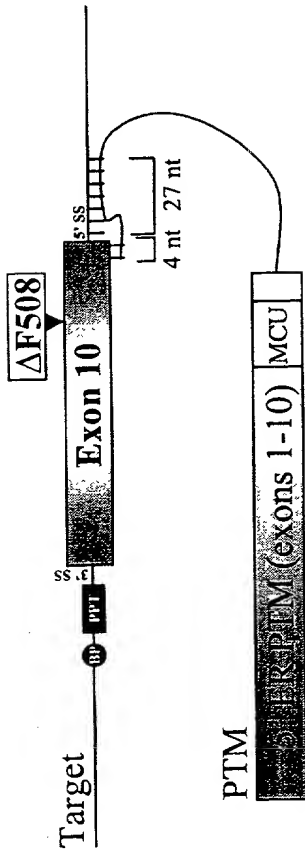
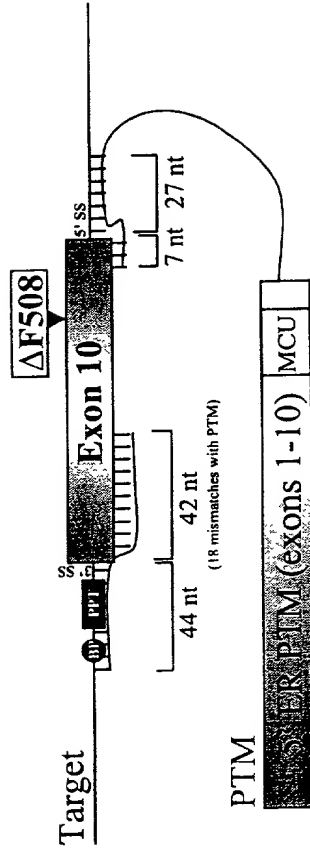


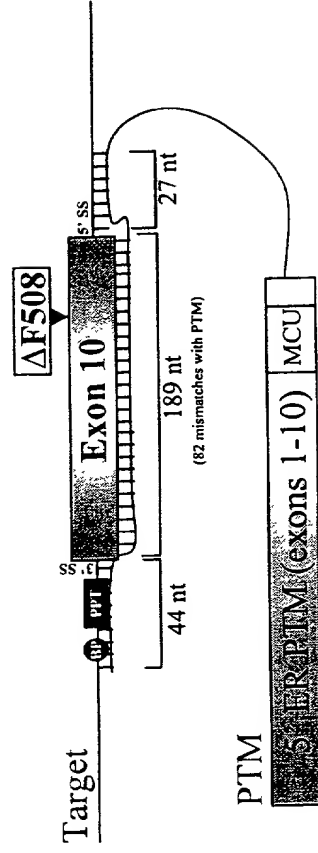
Figure 33



PTM with a short binding domain masking a single splice site in a mini-gene target.

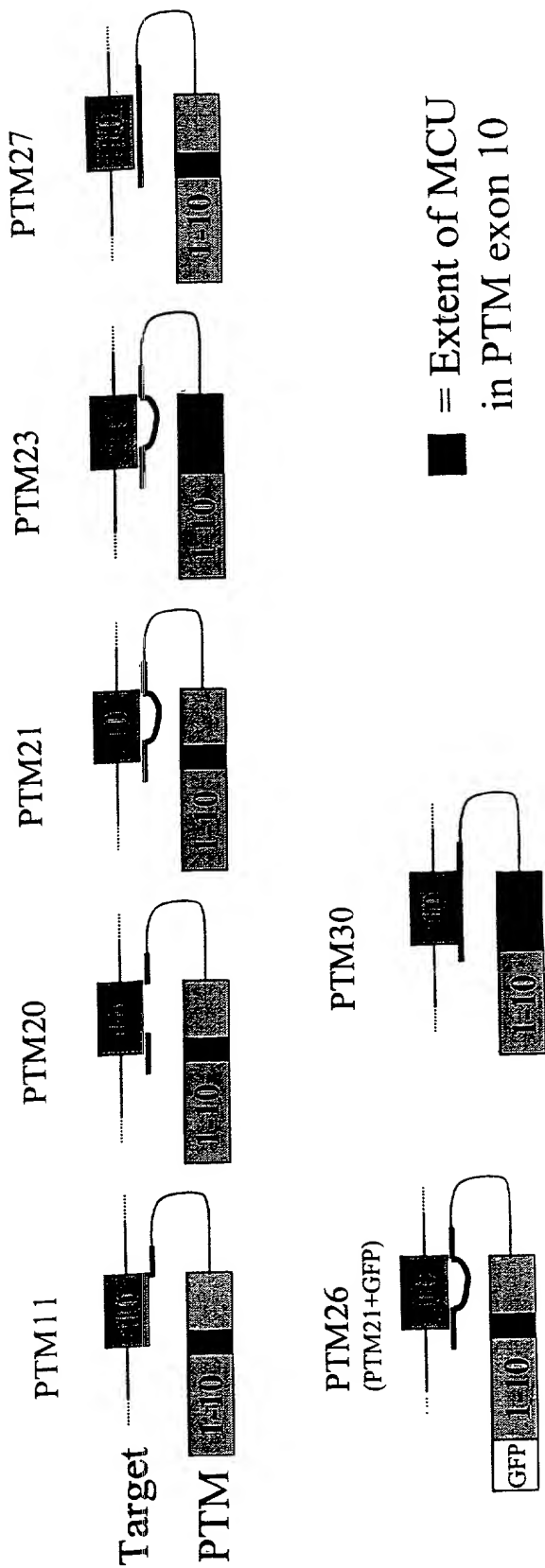


PTM with a long binding domain masking two splice sites in a mini-gene target.



PTM with a long binding domain masking two splice sites and the whole of exon 10 in a mini-gene target.

Figure 34



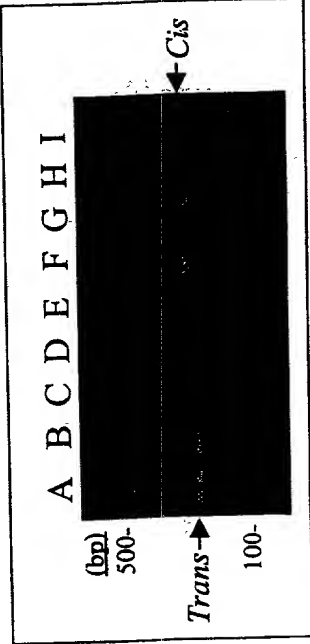
MCU in exon 10 of PTM
88 of 192 (46%) bases in PTM exon 10 are not complementary to its binding domain.

ACGAGCTTGCTCATGATGATGGCGAGTTAGAACCAAGTGAAAGGCAAGATCAAACATTCCG
GCCGCATCAGCTTTTCAGCCAATTCAGTTGGATCATGCCCGGTACCATCAAGGAGAACATAAT
CTTCGGCGTCAGTTACGACGAGTACCGCTATCGCTCGGTGATTAAGGCCTGTCAGTTGGAGGAG

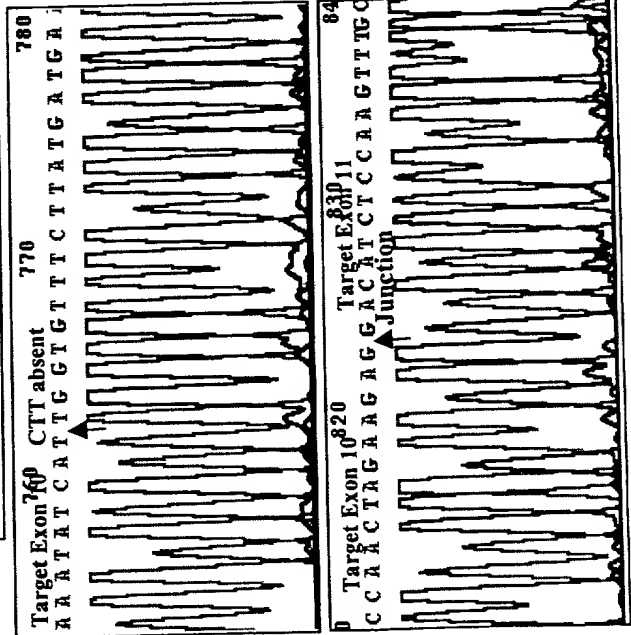
Figure 35

Sheet 43 of 58

INTRON



A.
Cis-spliced product
[Primers CF1 + CF111]



B.
Trans-spliced product
[Primers CF93 + CF111]

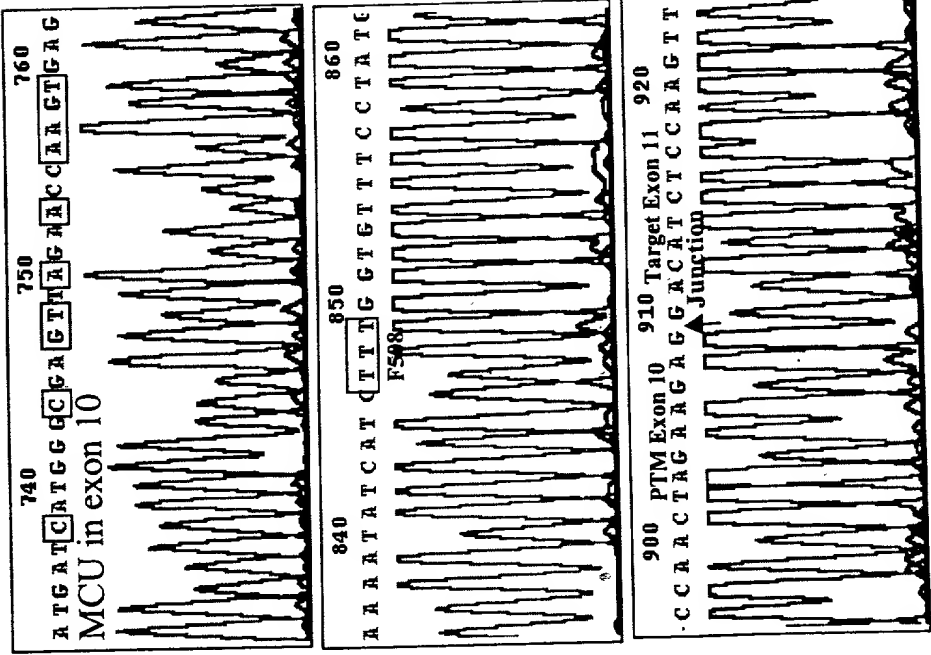


Figure 36

Sheet 44 of 58

A

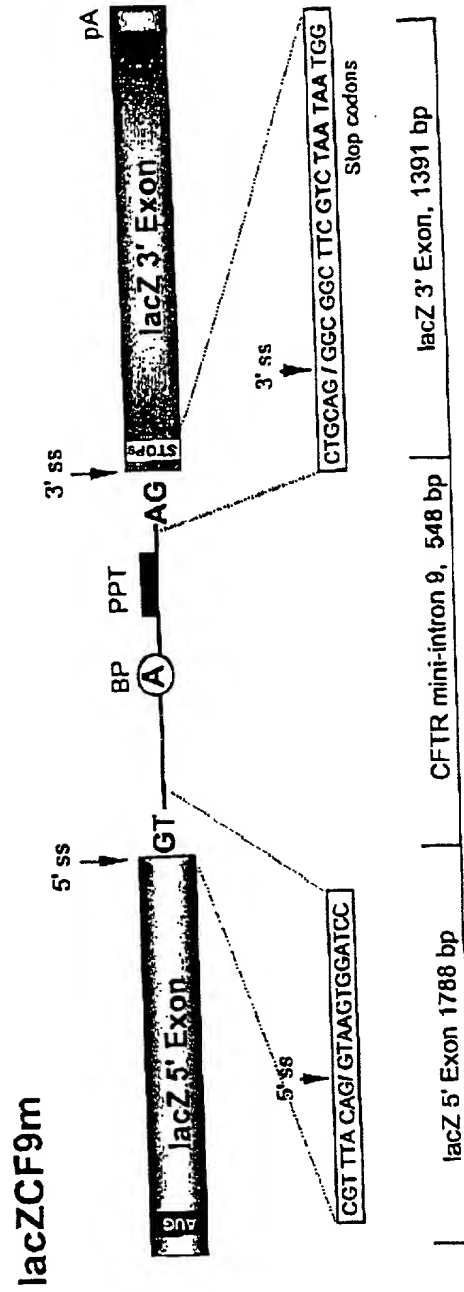


Figure 37 A

Sheet 45 of 58

B

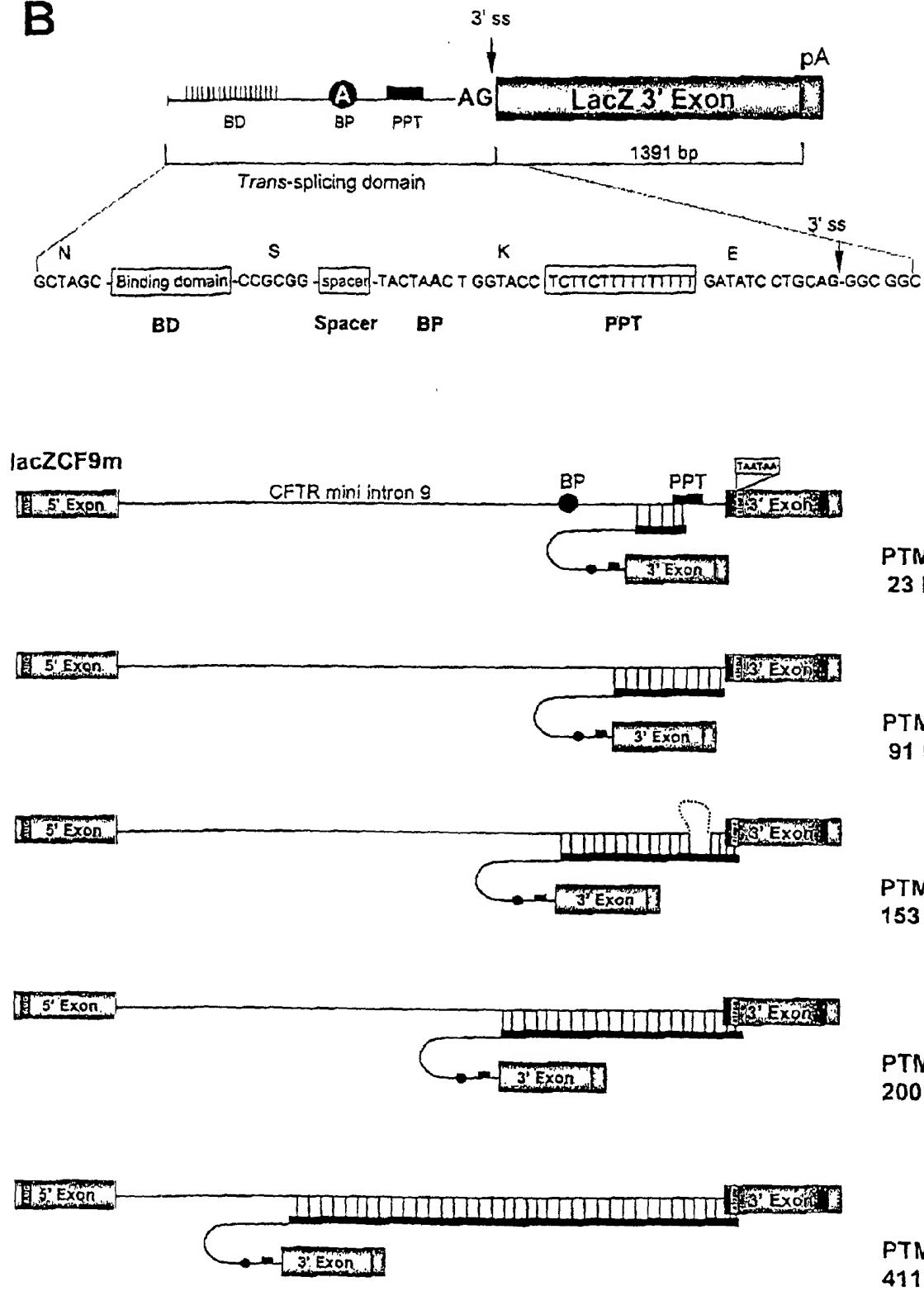


Figure 37B

Sheet 46 of 58

C

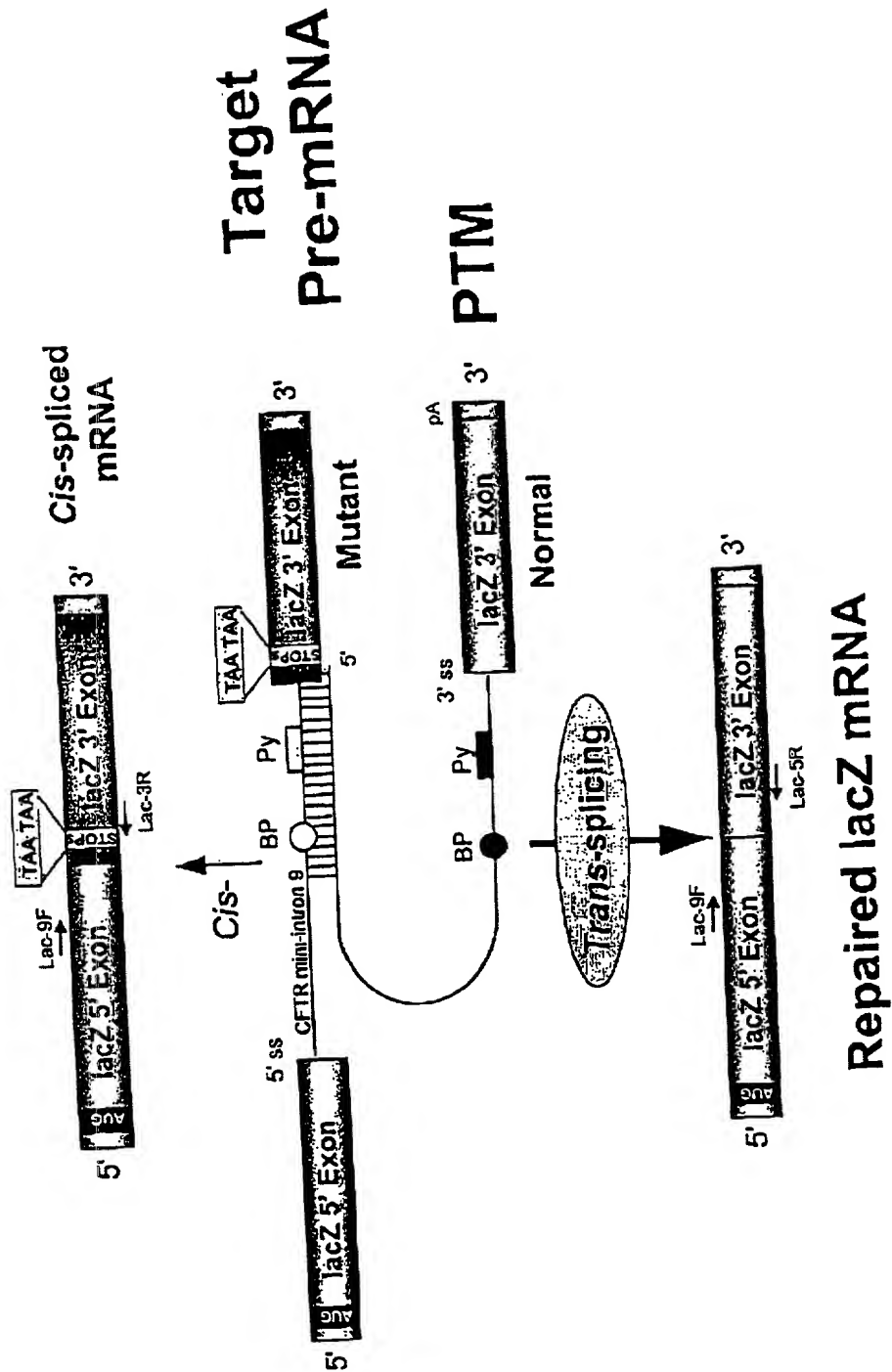


Figure 37C

Target
Pre-mRNA

PTM

Repaired lacZ mRNA

Sheet 47 of 58

A

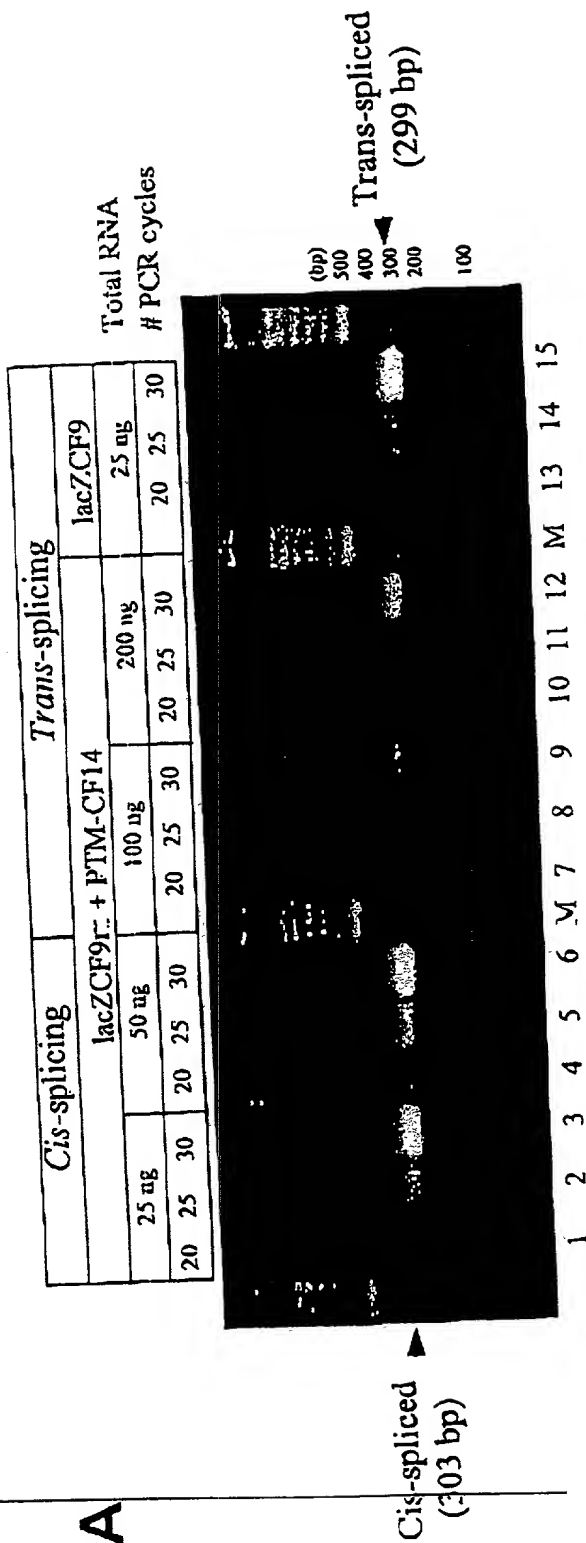
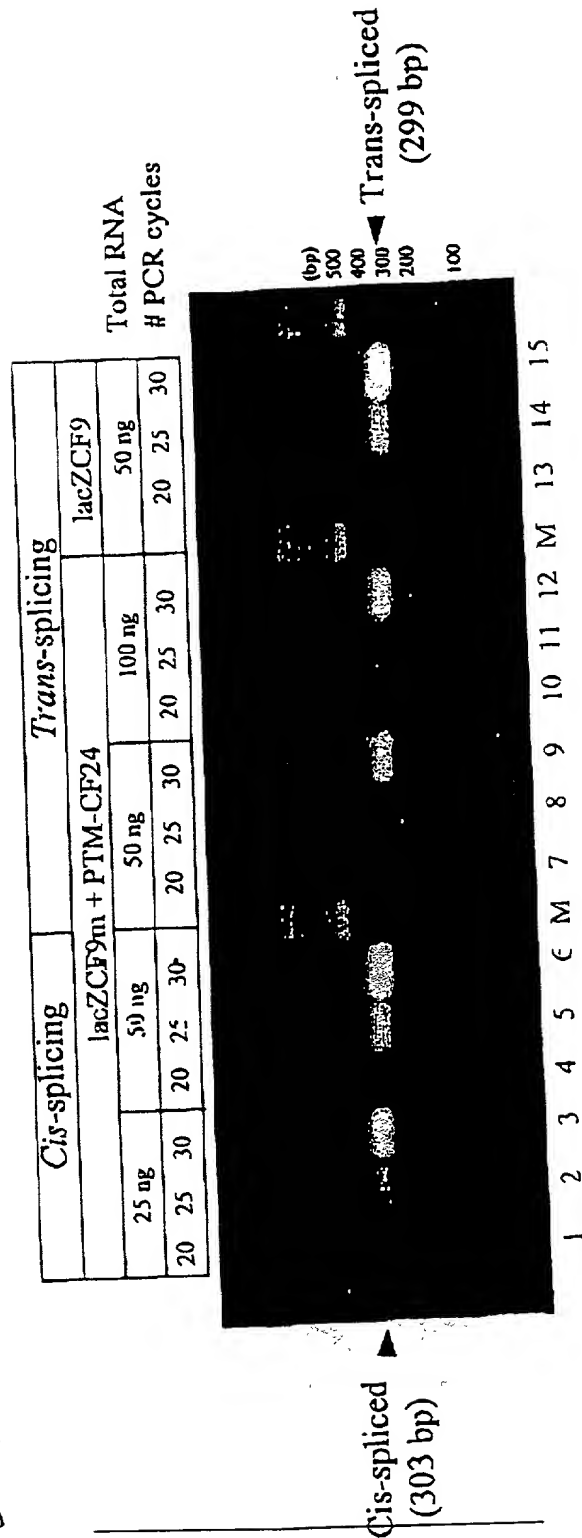


Figure 38 A



about 48 of 58

B

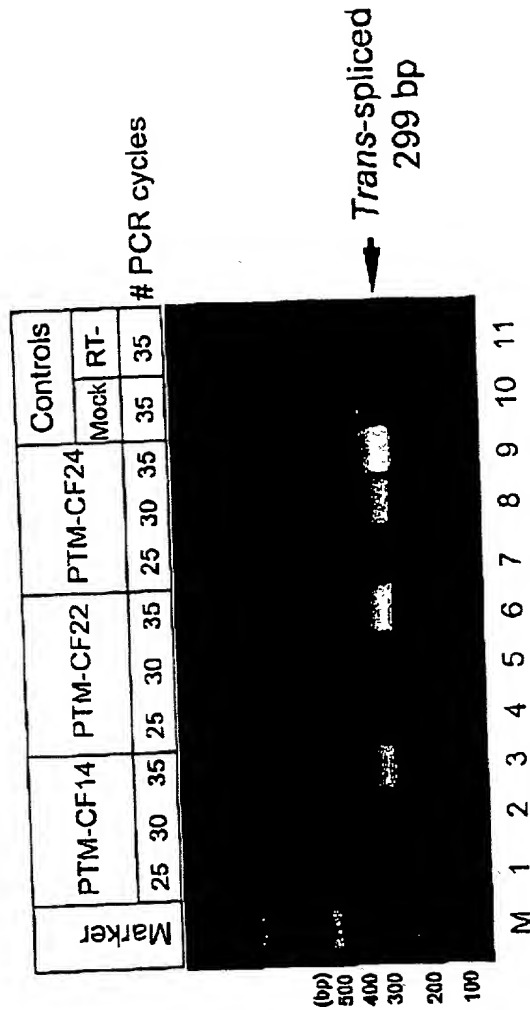


Figure 38B

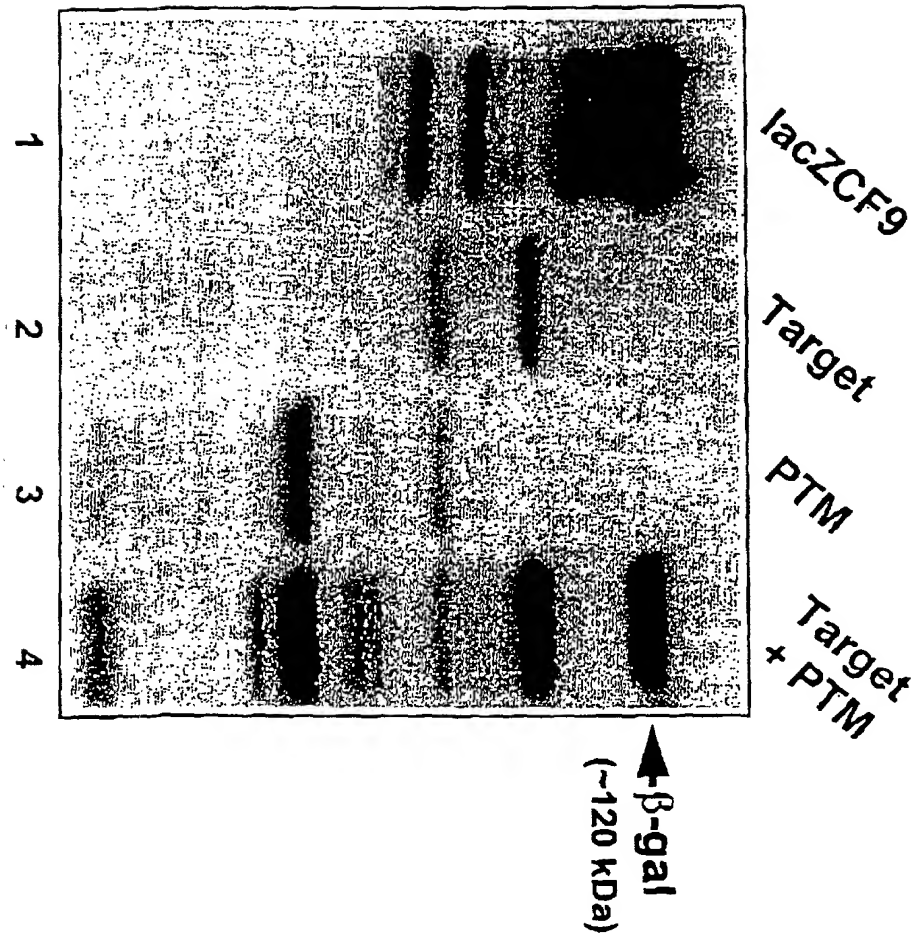


Figure 39

Figure 40A

(a)

(b)

החלטת הוועדה

Aut 51 of 58

B

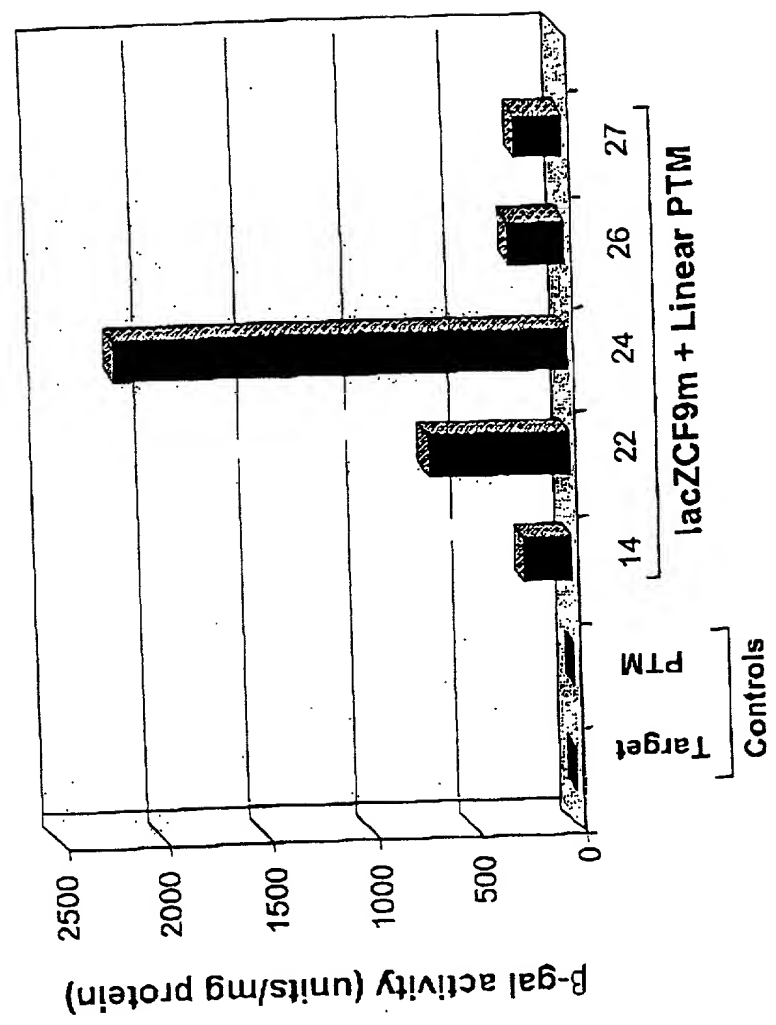


Figure 40B

Sheet 52 of 58

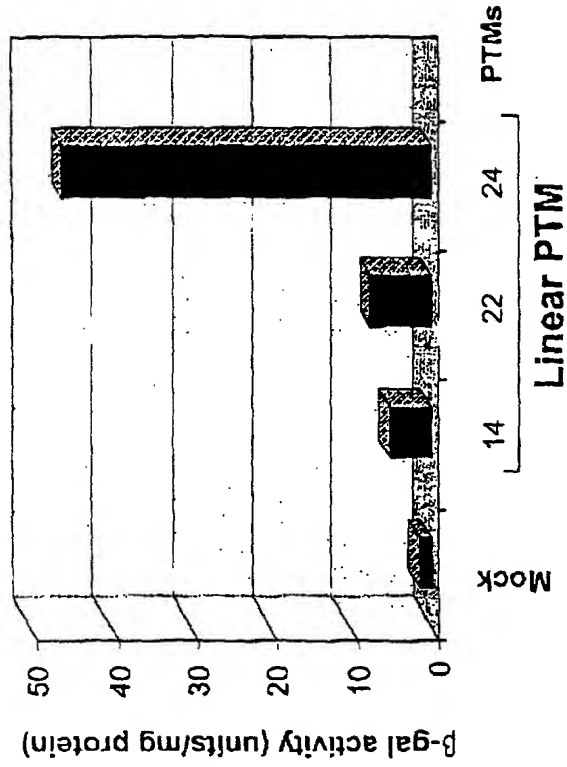


Figure 40C

C

9196862129

A

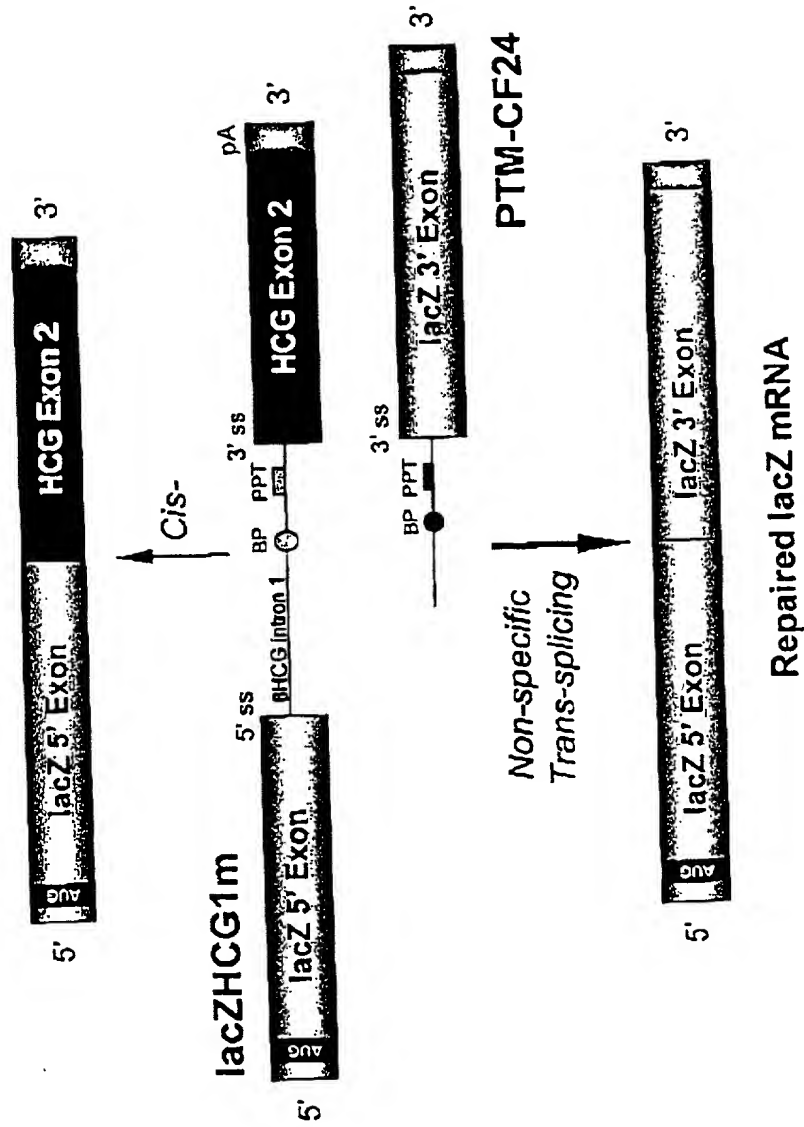


Figure 41A

Sheet 53 of 58

B

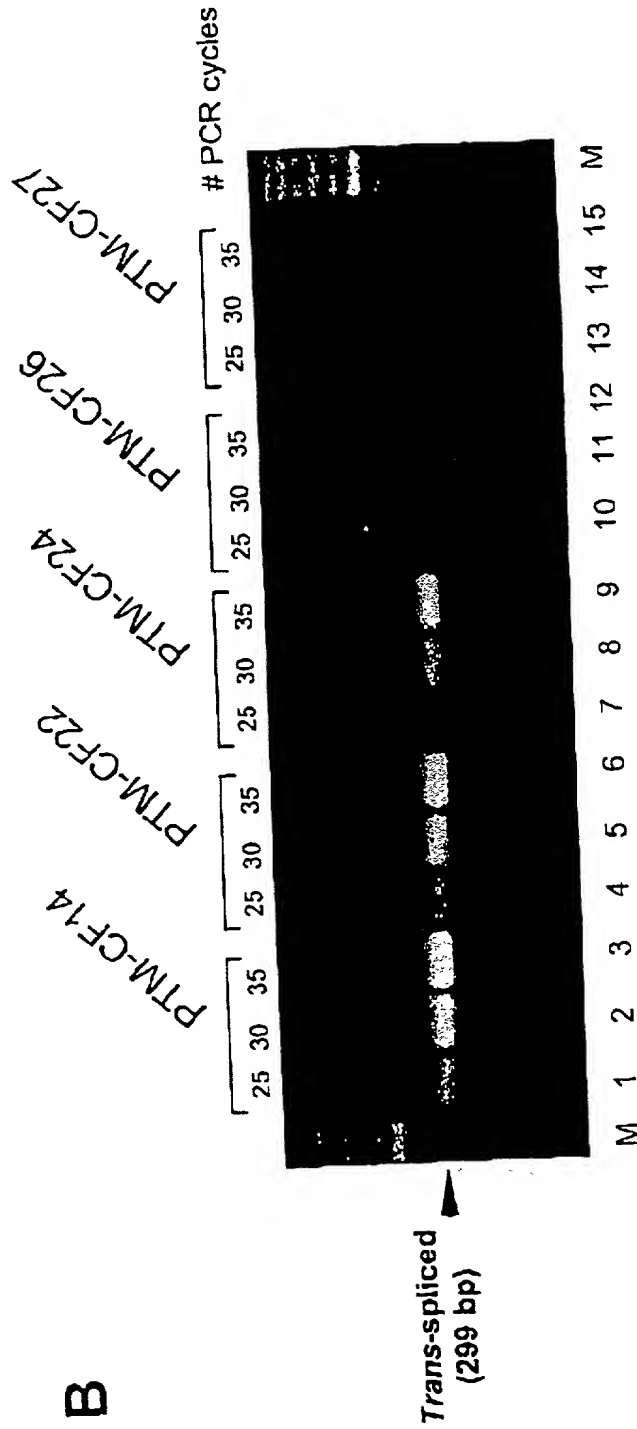


Figure 4B

Sheet 54 of 58

Sheet 55 of 58

C

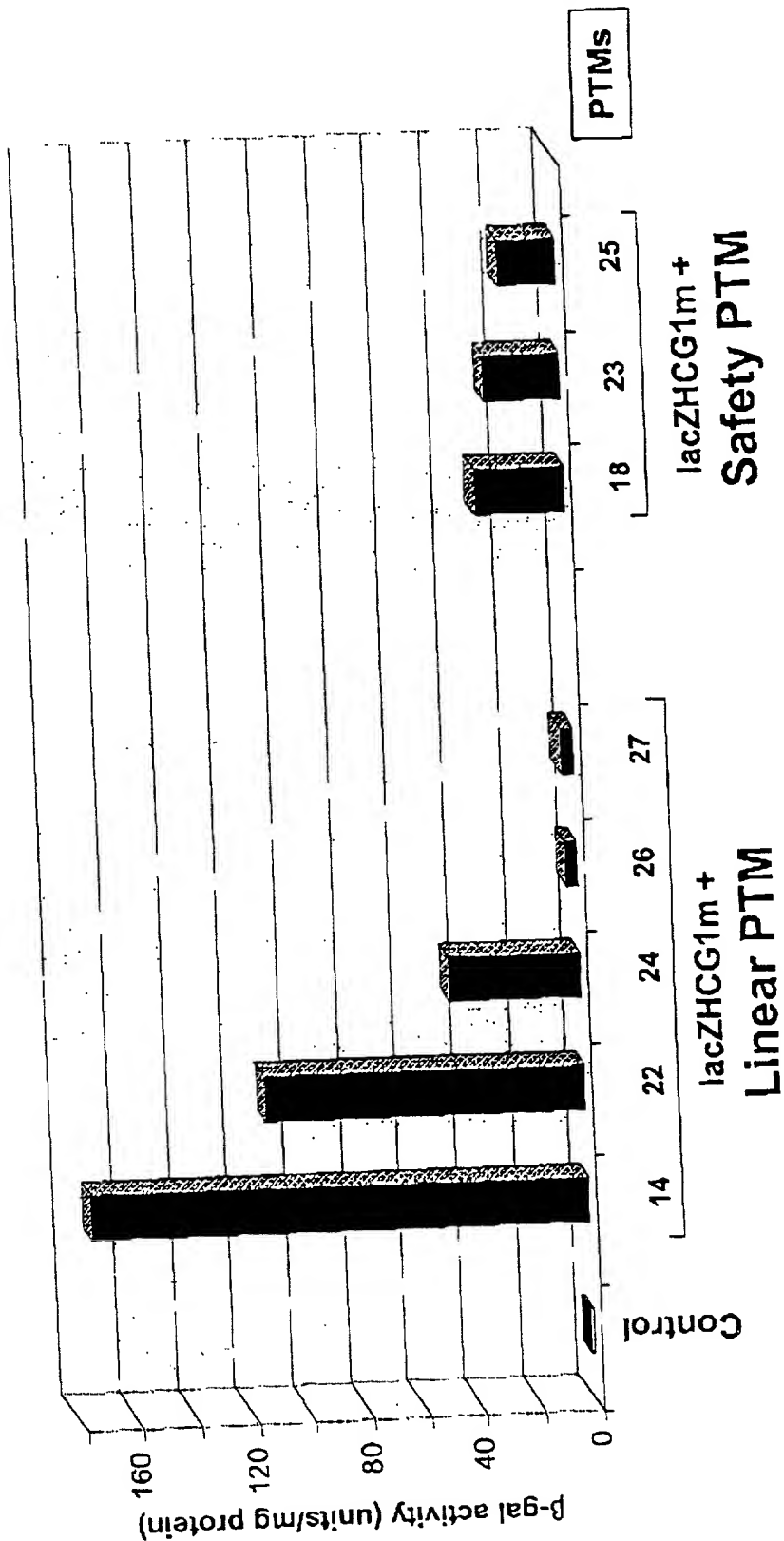


Figure 41C

Sheet 56 of 58

Exons 1-10

ATGCAGAGGTTCGCCTCTGGAAAAGGCCAGCGTTGTCTCCAAACTTTTTTTCAGCTGGACCAGACCAATTTTGAGGAAAAG
GATACAGACAGCGCCTGGAATTGTCAGACATATACCAAATCCCTTCTGTTGATTCTGCTGACAATCTATCTGAAAAATT
GGAAAGAGAATGGGATAGAGAGCTGGCTTCAAAGAAAAATCCTAAACTCATTAAATGCCCTTCGGCGATGTTTTTCTGG
AGATTTATGTTCTATGGAATCTTTTTATATTTAGGGGAAGTCACCAAAGCAGTACAGCCTCTCTTACTGGGAAGAATCA
TAGCTTCTCTATGACCCGGATAACAAGGAGGAACGCTCTATCGCGATTTATCTAGGCATAGGCTTATGCCTTCTCTTTAT
TGTGAGGACACTGCTCCTACACCCAGCCATTTTTGGCCTTCATCACATTGGAATGCAGATGAGAATAGCTATGTTTAGT
TTGATTTATAAGAAGACTTTAAAGCTGTCAAGCCGTGTTCTAGATAAAAATAAGTATTGGACAACCTTGTAGTCTCCTTT
CCAACAACCTGAACAAATTTGATGAAGGACTTGCAATTGGCACATTTTCGTGTGGATCGCTCCTTTGCAAGTGGCACTCCT
CATGGGGCTAATCTGGGAGTTGTTACAGGCGTCTGCCTTCTGTGGACTTGGTTTCTGTAGTCTTGCCTTTTTTCAG
GCTGGGCTAGGGAGAATGATGATGAAGTACAGAGATCAGAGAGCTGGGAAGATCAGTGAAAGACTTGTGATTACCTCAG
AAATGATCGAGAACATCCAATCTGTTAAGGCATACCTGCTGGGAAGAAGCAATGGAATAATGATTGAAAACCTTAAGACA
AACAGAACTGAAACTGACTCGGAAGGCAGCCTATGTGAGATACTTCAATAGCTCAGCCTTCTTCTCTCAGGGTCTTTT
GTGGTGTTTTTATCTGTGCTTCCCTATGCACTAATCAAAGGAATCATCCTCCGAAAAATTTACCACCATCTCATTCT
GCATTGTTCTGCGCATGGCGGTCACTCGGCAATTTCCCTGGGCTGTACAAACATGGTATGACTCTCTTGGAGCAATAAA
CAAAATACAGGATTTCTTACAAAAGCAAGAATATAAGACATTGGAATATAACTTAACGACTACAGAAGTAGTGATGGAG
AATGTAACAGCCTTCTGGGAGGAGGATTTGGGGAATTATTTGAGAAAGCAAAACAAACAATAACAATAGAAAACTT
CTAATGGTGATGACAGCCTCTTCTTCAAGTAATTTCTCACTTCTTGGTACTCCTGTCTGAAAGATATTAATTTCAAGAT
AGAAAGAGGACAGTTGTTGGCGGTTGCTGGATCCACTGGAGCAGGCAAGACGAGCTTGCTCATGATGATCATGGGCGAG
TTAGAACCAAGTGAAGGCAAGATCAAACATTCCGGCCGCATCAGCTTTTGCAGCCAATTCAGTTGGATCATGCCCGGTA
CCATCAAGGAGAACATAATCTTCGGCGTCAGTTACGACGAGTACCGCTATCGCTCGGTGATTAAGGCCTGTCAGTTGGA
GGAG

Trans-splicing domain

GTAAGATATCACCGATATGTGTCTAACCTGATTCCGGCCTTCGATACGCTAAGATCCACCGG
TCAAAAAGTTTTACATAATTTCTTACCTCTTCTGAATTCATGCTTTGATGACGCTTCTGTATCTATATTCATCATTG
GAAACACCAATGATATTTCTTTAATGGTGCCTGGCATAATCCTGGAAAACTGATAACACAATGAAATCTTCCACTGT
GCTTAATTTTACCCTCTGAATTCTCCATTTCTCCATAATCATATTACAACCTGAACCTCTGGAATAAAACCCATCATT
ATTAACCTCATTATCAAAATCAGCT

Figure 42

153 bp PTM24 Binding Domain:

Nhe I

153 bp BD underlined

GCTAGC - AATAATGACGAAGCCGCCCTCAGCTCAGGATTCACCTTGCCCTCCAATTATCATCCTAAGCAGAAGTGTATA

TTCTTATTGTAAAGATTCTATTAACTCATTTGATTCAAAATATTTAAATACTTCCTGTTTCACCTACTCTGCTATGC

Sac II

AC-CCGCCG

Figure 43A

Sheet 58 of 58

Trans-splicing domain

AATAATGACGAAGCCGCCCTCACGCTCAGGATTCACTTGCCCTCCAATTATCATCCTAAGCAGAAGTGTATATTCTTA
TTTGTAAGATTCTATTAACTCATTTGATTCAAAATATTTAAATACTTCCTGTTTCACCTACTCTGCTATGCACCCGC
GGAACATTATTATAACGTTGCTCGAATACTAAGTGTACCTCTTCTTTTTTTTTTGATATCCTGCAG

Exons 10-24

ACTTCACTTCTAATGATGATTATGGGAGAAGTGGAGCCTTCAGAGGGTAAAATTAAGCACAGTGAAGAATTCATTCT
GTTCTCAGTTTTCCTGGATTATGCCTGGCACCATTAAAGAAAATATCATCTTTGGTGTTCCTATGATGAATATAGATA
CAGAAGCGTCATCAAAGCATGCCAACTAGAAGAGGACATCTCCAAGTTTGCAGAGAAAGACAATATAGTTCTTGGAGAA
GGTGAATCAGACTGAGTGGAGGTCAACGAGCAAGAATTTCTTTAGCAAGAGCAGTATACAAAGATGCTGATTGTATT
TATTAGACTCTCCTTTTGATACCTAGATGTTTTAACAGAAAAAGAAATATTTGAAAGCTGTGTCTGTAAACTGATGGC
TAACAAAAGTAGGATTTTGGTCACTTCTAAAATGGAACATTTAAAGAAAGCTGACAAAATATTAATTTTGCATGAAGGT
AGCAGCTATTTTATGGGACATTTTCAGAACTCCAAAATCTACAGCCAGACTTTAGCTCAAAACTCATGGGATGTGATT
CTTTTCGACCAATTTAGTGCAGAAAGAAGAAATTCATCTTAAGTGTGAGACCTTACACCGTTTCTCATTAGAAGGAGATGC
TCCTGTCTCCTGGACAGAAACAAAAACAATCTTTTAAACAGACTGGAGAGTTTGGGGAAAAAGGAAGAATCTATT
CTCAATCCAATCAACTCTATACGAAAATTTTCCATTGTGCAAAAGACTCCCTTACAAATGAATGGCATCGAAGAGGATT
CTGATGAGCCTTTAGAGAGAAGGCTGTCTTAGTACCAGATTCTGAGCAGGGAGAGGCGATACTGCCTCGCATCAGCGT
GATCAGCACTGGCCCCACGCTTCAGGCACGAAGGAGGAGTCTGTCTGAACCTGATGACACACTCAGTTAACCAAGGT
CAGAACATTCACCGAAAGACAACAGCATCCACACGAAAAGTGTCACTGGCCCCCTCAGGCAAACTTGACTGAAGTGGATA
TATATTCAAGAAGGTTATCTCAAGAACTGGCTTGGAATAAGTGAAGAAATTAACGAAGAAGACTTAAAGGAGTGCTT
TTTTGATGATATGGAGAGCATACCAGCAGTGACTACATGGAACACATACCTTCGATATATTACTGTCCACAAGAGCTTA
ATTTTTGTGCTAATTTGGTGCTTAGTAATTTTTCTGGCAGAGGTGGCTGCTTCTTTGGTGTGTGCTGTGGCTCCTTGGAA
ACACTCCTCTTCAAGACAAAGGGAATAGTACTCATAGTAGAAATAACAGCTATGCAGTGATTATCACCAGCACCAGTTC
GTATTATGTGTTTTACATTTACGTGGGAGTAGCCGACACTTTGCTTGTATGGGATTCTTCAGAGGTCTACCACTGGTG
CATACTTAATCAGAGTGTGAAAAATTTTACACCACAAAATGTTACATTCTGTTCTTCAAGCACCTATGTCAACCCCTCA
ACACGTTGAAAGCAGGTGGGATTCTTAATAGATTCTCCAAAGATATAGCAATTTTGGATGACCTTCTGCCTCTTACCAT
ATTTGACTTTCATCCAGTTGTTATTAATTTGTGATTGGAGCTATAGCAGTTGTGCGAGTTTACAACCTACATCTTTGTT
GCAACAGTGCCAGTGATAGTGGCTTTTATTATGTTGAGAGCATATTTCTCCAAACCTCACAGCAACTCAAACAAGTGG
AATCTGAAGGCAGGAGTCCAATTTTCACTCATCTTGTTACAAGCTTAAAGGACTATGGACACTTCGTGCCTTCGGACG
GCAGCCTTACTTTGAAACTCTGTTCACAAAGCTCTGAATTTACATACTGCCAAGTGGTCTTGTACCTGTCAACACTG
CGCTGGTTCCAAATGAGAATAGAAATGATTTTTGTCTATCTTCTTCACTGCTGTTACCTTCATTTCCATTTTAAACAACAG
GAGAAGGAGAAGGAAGAGTTGGTATTATCCTGACTTTAGCCATGAATATCATGAGTACATTGCAGTGGGCTGTAACTC
CAGCATAGATGTGGATAGCTTGATGCGATCTGTGAGCCGAGTCTTTAAGTTTATTGACATGCCAACAGAAGGTAAACCT
ACCAAGTCAACCAACCATACAAGAAATGGCCAACTCTCGAAAGTTATGATTATTGAGAATTCACACGTGAAGAAAGATG
ACATCTGGCCCTCAGGGGGCCAAATGACTGTCAAAGATCTCACAGCAAAATACACAGAAGGTGGAAATGCCATATTAGA
GAACATTTCTCTCAATAAGTCTTGGCCAGAGGGTGGGCTCTTGGGAAGAACTGGATCAGGGAAGAGTACTTTGTTA
TCAGCTTTTTTGGAGACTACTGAACACTGAAGGAGAAATCCAGATCGATGGTGTGTCTTGGGATTCAATAACTTTGCAAC
AGTGGAGGAAAGCCTTTGGAGTGATACCACAGAAAGTATTTATTTTTCTGGAACATTTAGAAAAAACTTGGATCCCTA
TGAACAGTGGAGTGATCAAGAAATATGGAAGTTGCAGATGAGGTGGGCTCAGATCTGTGATAGAACAGTTTCTGGG
AAGCTTGACTTTGTCTTGTGGATGGGGCTGTGTCTTAAGCCATGGCCACAAGCAGTTGATGTCTTGGCTAGATCTG
TTCTCAGTAAGGCGAAGATCTTGCTGCTTGATGAACCCAGTGCTCATTGGATCCAGTAACATACCAATAATTAGAAG
AACTCTAAAAAAGCATTGTCTGATTGCACAGTAATCTCTGTGAACACAGGATAGAAGCAATGCTGGAATGCCAACAA
TTTTTGGTCATAGAAGAGAAACAAAGTGCAGCAGTACGATTCCATCCAGAACTGCTGAACGAGAGGAGCCTCTTCCGGC
AAGCCATCAGCCCCCTCCGACAGGGTGAAGCTTTTCCCCACCGAACTCAAGCAAGTGCAAGTCTAAGCCCCAGATTGC
Histidine tag Stop
TGCTCTGAAAGAGGAGACAGAAGAAGAGGTGCAAGATACAAGGCTTCATCATCATCATCATCATTAG

Figure 43B